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STUDIES OF THE THYROID APPARATUS. XXXV. THE RÔLE OF THE THYROID APPARATUS IN THE GROWTH OF THE ADRENALS

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INTRODUCTION

This paper deals with the growth in weight of the adrenal glands subsequent to thyroid and parathyroid removal from male and female albino rats at the ages of 23, 30, 50, 65, 75 and 100 days. The scope of the investigation and the procedure, methods of analysis and control have been given in previous papers. Suffice it to state that the animals were allowed to grow after the glandular extirpations until 150 days of age, when the various organs were removed and their weights compared with those of their controls. The method of computation leading to an evaluation of the respective rates of growth is given in the thirty-first paper of the series (1).

The mean observed weights of the adrenals of the several groups together with their probable errors are given in Table 1.

From these are computed the percentage increments and the percentage rate of growth of the tests in terms of that of their controls. These values are given in Table 2. It is easier to get an idea of general relations from charts than from tables of figures. I have, therefore, so arranged the percentage rate of growth of the tests in terms of that of their controls in a series of charts, that a visual comparison of the significant features may be had. The flat black columns represent control growth, while the outline columns give that of the tests. The significant contributions found in the literature up to 1923 have been given in two earlier papers (2), (3), and will not be referred to here unless some special need arises. For the reasons given elsewhere the growth response to thyro-parathyroidectomy will be interpreted in terms of thyroid deficiency alone (4).

THE EFFECT OF THYROID DEFICIENCY

On Chart 1A and B the growth of the adrenals is compared with that of the body in weight after thyroid removal at the stated ages. It is seen that the initiation of a thyroid deficiency at any time before the pubertal adjustment is in full swing (65 days of age) brings about a retardation in adrenal growth which parallels in direction the change in body weight with change in age at time of thyroid removal. Once the surge in incretory function of the gonads is established and the animal becomes capable of reproduction, the correlation between adrenal response and body weight response is destroyed.

It is, therefore, clear that up to the time when the pubertal development throws into the organism the new factors of influence emanating from the incretory products of the gonads, the growth processes of the adrenals are fundamentally like those of the body as a whole with respect to their reaction to thyroid deficiency. On the basis of the reasoning outlined in previous papers (5), (6), the conclusion is justified that before puberty the growth of the adrenals is not specifically related to thyroid activity.

The factors which come into play at puberty disrupt the simple dependence of the adrenals on the effectiveness of the growth processes of the body as a whole, and turn the organ over to the thyroid. As a result growth is not only stopped, but it is even rendered retrogressive: weight is actually lost

instead of gained. From which the conclusion seems necessary that the growth of the adrenals in the sexually mature albino rat is specifically dependent on thyroid function. Strong support for this conclusion is had from the fact that the adrenal is the only incretory gland which shows a specific positive weight association with the thyroid in the adult rat (7). The coefficient of correlation is 0.355 ± 0.059 in the male and 0.193 ± 0.065 in the female, when the general factors for organ size carried by the body weight and the influences assumed to be exerted by the other glands of internal secretion are held constant by the method of partial correlation.

Now an increase in the degree of growth retardation of the gonads in both sexes occurs when the thyroid is removed in late and post-pubertal stages of development (6). It can be assumed that this means a diminution in incretory function. Moreover the adrenals are positively and specifically correlated in weight with the gonads. The fifth order coefficient is 0.170 ± 0.056 in the male and 0.453 ± 0.036 in the female. This indicates a specific incretory relationship. Riddle (8) has recently demonstrated a similar relation in female pigeons. Hence it is clear that the pubertal and post-pubertal reaction of the adrenals to thyroid deficiency is due to the retardation of gonadal development as well as to the specific adrenal-thyroid incretory relationship. Further support for this idea is had from the fact that the daily percentage rate of growth of the adrenals in the normal rat drops abruptly between the ages of 65 and 75 days to the level characteristic of the adult animal. The change is from 4.0 to 0.2 in the male, and from 5.2 to 1.0 in the female. While this great diminution in growth capacity is of itself a factor in the reaction of the adrenals to thyroid deficiency, the fact that it occurs at puberty justifies the assumption that it is dependent in part at least on gonadal incretory activity, and hence that these are conditioners of the response to thyroid removal.

From Charts 3A and 1A and B it is seen that the growth reaction of the adrenal to thyroid deficiency is both absolutely and relatively with respect to body weight greater in the female than in the male. The fact that this relation is the opposite of that to be expected from the percentage increment in the control rats (Table 2), indicates either specificity of adrenal-

TABLE 1

The Observed Mean Weights of the Adrenals of the Several Groups of Rats

Age Series	THYROPARATHYROIDECTOMIZED					PARATHYROIDECTOMIZED			
	At Beginning			At End (150 days)		At Beginning		At End (150 days)	
	Ref. Contl.	Contl.	Test	Control	Test	Contl.	Test	Control	Test
	MALES								
23	gm. 0.0067±0.0003	gm. 0.0070	gm. 0.0068	gm. 0.0256±0.0005	gm. 0.0235±0.0007	gm. 0.0068	gm. 0.0066	gm. 0.0264±0.0007	gm. 0.0218±0.0006
30	0.0093±0.0003	0.0091	0.0090	0.0263±0.0007	0.0206±0.0008	0.0092	0.0092	0.0272±0.0007	0.0238±0.0005
50	0.0141±0.0002	0.0137	0.0136	0.0250±0.0008	0.0185±0.0009	0.0138	0.0135	0.0255±0.0007	0.0225±0.0004
65	0.0220±0.0010	0.0209	0.0212	0.0264±0.0006	0.0200±0.0006	0.0213	0.0214	0.0256±0.0007	0.0205±0.0007
75	0.0224±0.0004	0.0215	0.0218	0.0254±0.0008	0.0216±0.0005	0.0219	0.0215	0.0256±0.0012	0.0186±0.0007
100	0.0227±0.0006	0.0224	0.0222	0.0243±0.0007	0.0192±0.0005	0.0224	0.0219	0.0243±0.0007	0.0214±0.0005
	FEMALES								
23	0.0073±0.0003	0.0071	0.0069	0.0475±0.0012	0.0269±0.0013	0.0073	0.0072	0.0483±0.0021	0.0337±0.0019
30	0.0083±0.0004	0.0082	0.0081	0.0441±0.0011	0.0234±0.0011	0.0083	0.0082	0.0453±0.0012	0.0353±0.0011
50	0.0147±0.0005	0.0141	0.0141	0.0385±0.0007	0.0215±0.0011	0.0147	0.0144	0.0401±0.0009	0.0284±0.0015
65	0.0264±0.0008	0.0257	0.0249	0.0388±0.0016	0.0248±0.0013	0.0258	0.0253	0.0382±0.0013	0.0290±0.0016
75	0.0294±0.0014	0.0285	0.0281	0.0401±0.0011	0.0262±0.0014	0.0279	0.0280	0.0426±0.0019	0.0291±0.0018
100	0.0368±0.0017	0.0356	0.0356	0.0415±0.0017	0.0240±0.0009	0.0356	0.0351	0.0415±0.0017	0.0320±0.0014

thyroid association or interference from some third factor (7). Since specificity of adrenal-thyroid relationship is not marked until gonad incretory activity is well in force, and since the degree of association between adrenal and ovary is greater than that between adrenal and testis, and since ovary growth is more retarded by thyroid deficiency than is testis growth, the conclusion must be made that the sex differential response of the adrenals is due to an interference on the part of the gonads.

TABLE 2

The Absolute and the Relative Percentage Rate of Growth of the Adrenals of the Several Groups

Age Series	MALES			FEMALES		
	Thyroparathyroidectomized Groups					
	Control	Test	T/C	Control	Test	T/C
	%	%	%	%	%	%
23	265.7	245.6	92.4	569.0	289.9	50.9
30	189.0	128.9	68.2	437.8	188.9	43.1
50	82.5	36.0	43.7	173.1	52.5	30.3
65	26.3	—5.7	—21.5	51.0	—0.4	—0.8
75	18.1	—0.9	—5.1	40.7	—6.8	—16.6
100	8.5	—13.5	—159.3	16.6	—32.6	—196.6
	Parathyroidectomized Groups					
23	288.2	230.3	79.9	561.6	368.1	65.5
30	195.7	158.7	81.1	445.8	330.5	74.1
50	84.8	66.7	78.6	172.8	97.2	56.3
65	20.2	—4.2	—20.9	48.1	14.6	30.4
75	16.9	—13.5	—79.9	52.7	3.9	7.5
100	8.5	—2.3	—26.9	16.6	—8.8	—53.3

* These figures are positive because control growth being retrogressive gives a percentage decrement instead of increment. The quotient of this into the percentage decrement of the tests is necessarily algebraically positive in sign. The values represent the relative degree of loss of weight, notwithstanding, and are so charted.

The degree of association of the adrenals with the body weight is higher in the adult female than in the male. The values for the coefficient of correlation are 0.552 ± 0.043 and 0.329 ± 0.055, respectively. This may be a factor in the greater

regression of adrenal weight in the thyroidless females of the 100 day old series (Chart 1A and B).

THE EFFECT OF PARATHYROID DEFICIENCY

From Chart 2A it is seen that the initiation of a parathyroid deficiency in the male at any time before the pubertal adjustment is in full swing (65 days of age) brings about a retardation of adrenal growth, which while less in degree, paral-

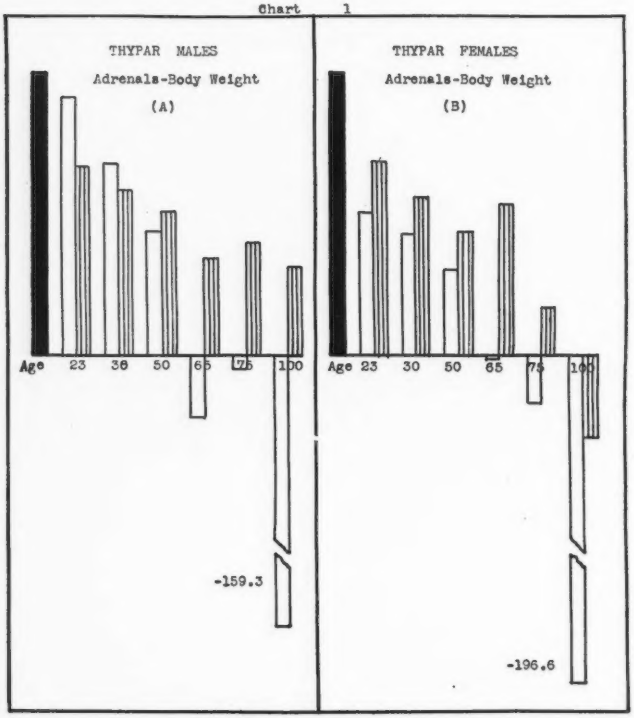

Chart 1. Comparing growth retardation of adrenals and body weight after thyroid removal from A—Males and B—Females at the stated ages. Flat black column represents control growth; outline columns, growth of the test animals. Order of sequence given by the superscript.

lels in direction that of the body weight with respect to the age difference in response. Once puberty is established, however, and the animal becomes capable of reproduction the associa-

tion is ruptured and the adrenal response is in no way comparable with that of the body. In the female, on the other hand (Chart 2B) the growth response of the adrenals, while greater in degree, is quite consistently parallel with that of the body in the direction of change with change in age at time of glandular removal.

The conclusion is that in general the growth processes of the adrenals of the female are similar to those of the body as a whole in their reaction to parathyroid deficiency and are sim-

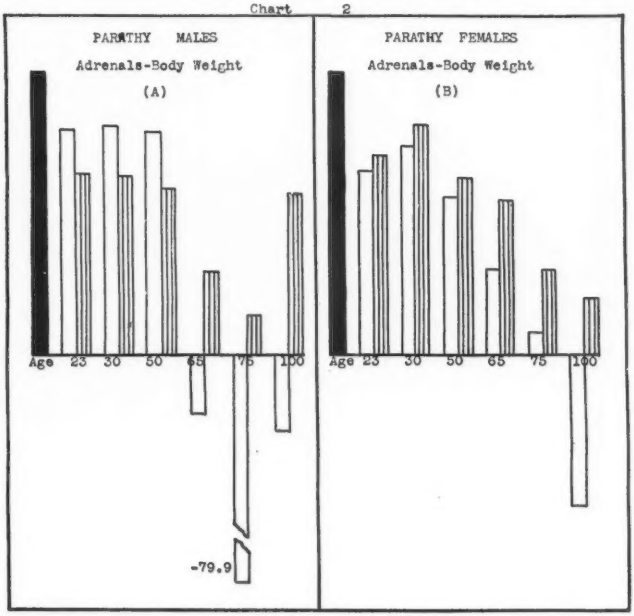

Chart 2. Comparing growth retardation of adrenals and body weight after parathyroid removal from A—Males and B—Females at the stated ages. Flat black column represents control growth; outline columns, growth of the test animals. Order of sequence given by the superscript.

ilarly interpretable, i. e., in terms of a lowered nutritional level (4). The regression of adrenal growth in the 100 day old series is like that which occurs in the case of the ovary (7). From what has gone before it is evident that the extra reaction of the former can be attributed to that of the latter. This raises the question as to whether the general concordance of adrenal re-

sponse with body weight response is due to its dependence on the effectiveness of the growth processes of the body as a whole, or whether it is due to the specific adrenal-ovary association, since the ovary also tends to follow the course of the body weight response. Inasmuch as the adrenal exhibits a closer parallelism than the ovary in this respect, I am inclined to believe that the

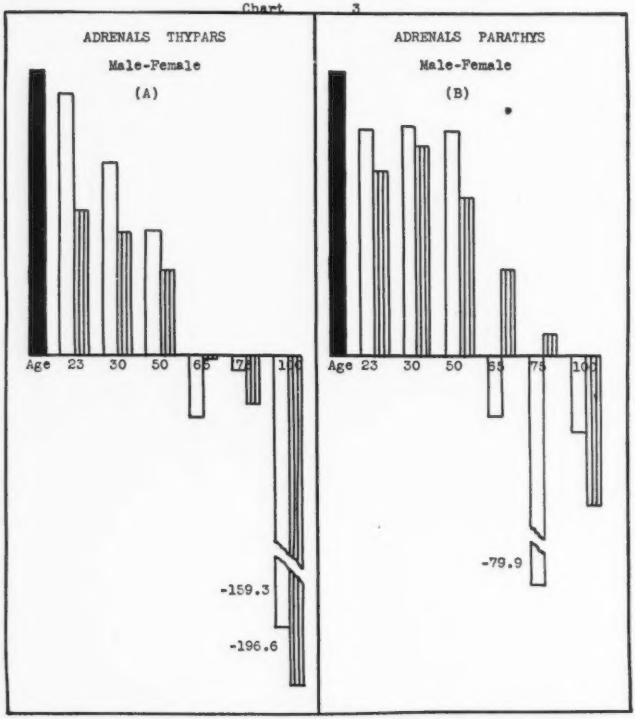

Chart 3. Sex comparison of adrenal response to A—Thyroid removal and B—Parathyroid removal at the stated ages. Flat black column represents control growth; outline columns, growth of the test animals. Order of sequence given by the superscript.

first interpretation is the more valid, at least up to the age of sexual maturity. At any rate it is clear that there is no specific relation between adrenal growth and parathyroid function.

In the case of the males the same conclusion holds true for groups from which the parathyroids are removed prior to the

pubertal surge. The marked reaction which occurs during and after this period shows conclusively that puberty conditions an increased sensitivity to parathyroid deficiency. Since the reaction of the adrenals is so markedly different from that of the body as a whole and of the parts so far studied, it would appear

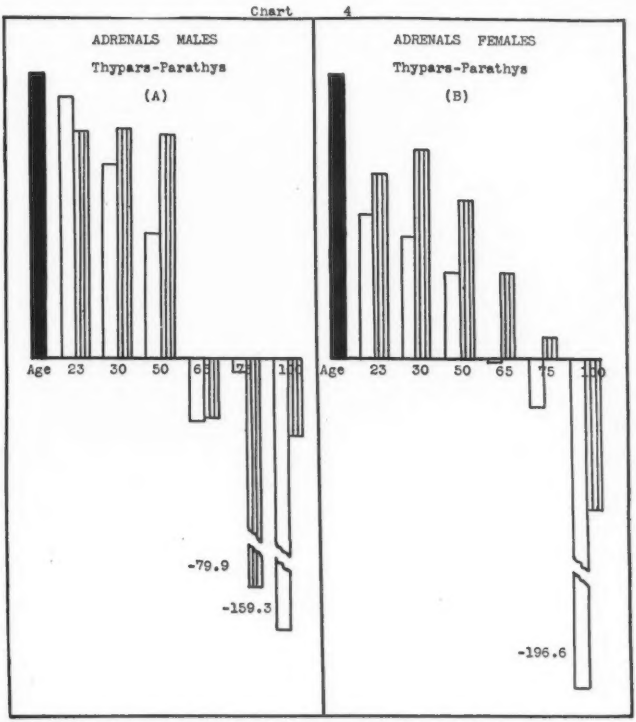

Chart 4. Comparing the effect of thyroid and parathyroid deficiency on adrenal growth in A—Male and B—Female albino rats. Flat black column represents control growth; outline columns, growth of the test animals. Order of sequence given in the superscript.

as if a certain degree of specificity is present. On the other hand, since it is puberty that brings this about, and since the pubertal changes are justly attributable to testicular incretory activities, it would also appear as if it is these changes and their influence on the body and not any specific adrenal-parathyroid incretory relation which is at the basis of the matter.

Perhaps the situation can best be expressed by saying that puberty in the male determines an increased sex-specific sensitivity of the adrenals to parathyroid deficiency. The effect of this is shown on Chart 3B.

From Chart 4B it is seen that the growth of the adrenals in the female is uniformly less disturbed by parathyroid than by thyroid deficiency. In the male the relations are inconstant (Chart 4A). In both sexes, however, the relative influence of the two types of glandular deficiency is the same in direction as that exhibited by the body in weight. Hence it can be concluded that these differences are essentially an expression of the dependency of the adrenals on the relative effectiveness of the growth processes of the body as a whole after thyroid and parathyroid removal.

Now a study of the course of growth in body weight subsequent to thyroid and parathyroid removal (4) led to the conclusion that the growth retardation which ensues is the result of a condition of essential undernutrition in both cases. The question therefore arises as to whether the growth of the adrenals in the thyroidless and parathyroidless rats is like that which occurs in inanition. While a study of the effects of undernutrition on the growth of the adrenals as given by Jackson (9) shows that these organs are markedly influenced by dietary deficiencies, the singular inconstancy of the results indicates that other factors beside that of diet are at work. True it is that an attempt has been made to evaluate the influence of age and sex, but only confusion has resulted since the number of animals used in any given group was too small and the experimental conditions too varied for accurate outcome. These preliminary explorations show that the matter deserves a carefully planned and systematic investigation.

The present state of the subject allows no definite correlation to be made between the adrenal response to dietary undernutrition and that resulting from thyroid or parathyroid deficiency. From Charts 1 and 2 it is clear that in the male the adrenals are less sensitive than the body as a whole to the uncomplicated nutritional deficiency originating from thyroid or parathyroid removal before puberty. Whether a similar sex difference will be found in cases of undernutrition due to dietary deficiency is yet to be determined. That such is possible is in-

dicated from the report of Jackson and Stewart (10) of a sex difference of like direction in rats underfed from three weeks to twenty weeks of age, and then refed for a time.

SUMMARY AND CONCLUSIONS

The outstanding facts brought out by this study are that the incretory relationship of the gonads with the adrenals is a major factor in determining the growth response of the adrenals to thyroid deficiency, and that puberty determines a sex-specific sensitivity of the adrenals to parathyroid deficiency in the male.

Other findings are:

1. The growth of the adrenals is not specifically related to thyroid activity before puberty.

2. The growth of the adrenals in the sexually mature albino rat is specifically related to thyroid activity.

3. The growth of the adrenals in the female rat is not specifically related to parathyroid activity.

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STUDIES OF THE THYROID APPARATUS. XXXVI. THE RÔLE OF THE THYROID APPARATUS IN THE GROWTH OF THE PANCREAS

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INTRODUCTION

This paper deals with the growth of the pancreas of male and female albino rats subsequent to thyroid and parathyroid removal at 23, 30, 50, 65, 75 and 100 days of age. Since preceding papers give the scope, methods and point of view of the investigation as a whole, none of the details need be given here. Attention is called to the fact, however, that the animals were 150 days of age at the time the terminal records were made. Table 1 contains the observed means and probable errors of the weights of the pancreas of the several groups. The computation of the initial "control" and "test" values is described elsewhere (1). Table 2 gives the percentage rate of growth calculated from the data in Table 1. It also gives the percentage rate of growth of the tests in terms of that of their controls (T/C). The comparisons are best made from charts. On these the flat black columns represent control growth (always 100 per cent), while the outline columns stand for test growth in terms of control. The order of sequence is given by the superscript. For the reasons given in another paper (2) the sequelæ of thyro-parathyroidectomy are interpreted in terms of the thyroid deficiency alone.

THE INFLUENCE OF THYROID DEFICIENCY

On Chart 1A and B the growth of the pancreas is compared with that of the body in weight after thyroid removal at the stated ages.

It is evident that the sensitivity of the pancreas is greater than that of the body as a whole; that this relatively greater sensitivity increases markedly at the onset of puberty, weight actually being lost instead of gained; and that the sensitivity of the

TABLE 1

The Observed Mean Weight of the Pancreas of the Several Groups.

Age Series	THYROPARATHYROIDECTOMIZED					PARATHYROIDECTOMIZED			
	At Beginning			At End (150 days)		At Beginning		At End (150 days)	
	Ref. Contl.	Contl.	Test	Control	Test	Contl.	Test	Control	Test
	MALES								
23	gm. 0.1296 ± 0.0046	gm. 0.1371	gm. 0.1333	gm. 0.8337 ± 0.0279	gm. 0.5549 ± 0.0231	gm. 0.1330	gm. 0.1292	gm. 0.8456 ± 0.0264	gm. 0.6216 ± 0.0175
30	0.2512 ± 0.0040	0.2458	0.2421	0.7736 ± 0.0230	0.4685 ± 0.0297	0.2495	0.2486	0.8332 ± 0.0267	0.7221 ± 0.0212
50	0.4114 ± 0.0146	0.4005	0.3973	0.8246 ± 0.0281	0.5014 ± 0.0147	0.4028	0.3947	0.8131 ± 0.0231	0.6594 ± 0.0276
65	0.5918 ± 0.0192	0.5631	0.5705	0.8149 ± 0.0164	0.5238 ± 0.0169	0.5729	0.5754	0.8400 ± 0.0142	0.6401 ± 0.0254
75	0.5909 ± 0.0180	0.5662	0.5740	0.8225 ± 0.0246	0.5384 ± 0.0200	0.5784	0.5669	0.8177 ± 0.0198	0.5487 ± 0.0240
100	0.6315 ± 0.0149	0.6229	0.6184	0.7622 ± 0.0205	0.5363 ± 0.0183	0.6229	0.6083	0.7622 ± 0.0205	0.7010 ± 0.0204
	FEMALES								
23	0.1379 ± 0.0058	0.1334	0.1308	0.7527 ± 0.0269	0.4232 ± 0.0261	0.1380	0.1364	0.7244 ± 0.0198	0.5644 ± 0.0217
30	0.2602 ± 0.0073	0.2557	0.2532	0.7071 ± 0.0218	0.4000 ± 0.0148	0.2604	0.2557	0.7378 ± 0.0212	0.6693 ± 0.0163
50	0.4168 ± 0.0092	0.4009	0.3998	0.6971 ± 0.0105	0.3909 ± 0.0161	0.4159	0.4095	0.7084 ± 0.0106	0.5800 ± 0.0137
65	0.5667 ± 0.0181	0.5513	0.5352	0.6483 ± 0.0248	0.4525 ± 0.0201	0.5537	0.5429	0.7058 ± 0.0167	0.5838 ± 0.0204
75	0.5676 ± 0.0158	0.5499	0.5424	0.6287 ± 0.0208	0.4308 ± 0.0166	0.5394	0.5404	0.6469 ± 0.0313	0.5457 ± 0.0308
100	0.6485 ± 0.0140	0.6280	0.6280	0.6914 ± 0.0210	0.4923 ± 0.0138	0.6280	0.6185	0.6914 ± 0.0210	0.6452 ± 0.0279

* "Beginning" values for "control" and "test" groups computed according to method referred to in text.

pancreas like that of the body progressively increases with the increase in age at the time of thyroid removal.

The picture as a whole gives the impression that the growth processes of the pancreas are more dependent upon the effectiveness of the growth processes of the body as a whole, than upon any specific relation to thyroid activity. This is consistent with the fact that the positive weight correlation which exists between pancreas and thyroid (0.339 ± 0.064 in the male, and 0.235 ± 0.070 in the female) in the adult rat is largely due to body size

TABLE 2

The Absolute and the Relative Rate of Growth of the Pancreas of the Several Groups.

Age Series	MALES			FEMALES		
	Thyroparathyroidectomized Groups					
	Control	Test	T/C	Control	Test	T/C
	%	%	%	%	%	%
23	508.1	316.3	62.2	464.2	223.6	48.2
30	214.7	93.5	43.6	176.5	58.0	32.8
50	105.9	26.2	24.7	73.9	—2.2	—3.0
65	44.7	—8.2	—18.3	17.6	—15.5	—87.8
75	45.3	—6.2	—13.7	14.3	—20.6	—143.6
100	22.4	—13.3	—59.4	10.1	—21.6	—214.0
	Parathyroidectomized Groups					
23	535.8	381.1	71.1	424.9	313.8	73.8
30	234.0	190.5	81.4	183.3	161.8	88.2
50	101.9	67.1	65.8	70.3	41.6	59.2
65	46.6	11.2	24.1	27.5	7.5	27.4
75	41.4	—3.2	—7.8	19.9	1.0	4.9
100	22.4	15.2	68.2	10.1	4.3	42.8

influence, for when this is held constant by the method of partial correlation the association is negligible (0.115 in the male and 0.049 in the female). Moreover, there is no specific weight correlation between the two organs when the assumed influences of the other incretory organs are collectively stabilized for. The 5th order coefficient is 0.063 in the male and —0.010 in the female.

The pubertal and post-pubertal atrophy of the pancreas would seem to indicate a direct conditioning of the pancreas response to thyroid deficiency by the incretory activity of the gonads. Opposed to this is the fact that the weight correlation between gonads and pancreas (0.403 ± 0.051 in the male and 0.374 ± 0.053 in the female) is largely dependent on body size, for when this is held constant by the method of partial correlation the degree of association drops to 0.083 in the male and 0.129 in the female. On the other hand, the difference in character of pancreas and body weight response in the 65, 75 and 100 day old series shows that a factor other than body size is effective. This can arise from no other organ than the gonads. But there is no significant direct weight association between pancreas and gonads in the sexually mature rat as shown by the coefficients of correlation just given, which are substantiated by the 5th order values (that for the male being 0.093 and that for the female —0.024). These facts lead to the inference that a third factor participates in the reaction. This factor is the adrenals.

It has been shown in an earlier paper (3) that although there is no general specific weight association between the several glands of internal secretion; there is, on the other hand, an indirect association which is mediated by the adrenals. Now the weight correlation between gonads and adrenals is not only somewhat independent of body size, but it is also independent of the influences assumed to be exerted by the other incretory organs. This is shown by the fact that the zero order values of 0.310 ± 0.056 in the male and 0.647 ± 0.036 in the female remain positive both when body weight is alone held constant (0.143 in the male and 0.494 in the female) and when in addition the thyroid, thymus, hypophysis and pancreas are collectively stabilized for by the method of partial correlation. The value for the male is 0.170 and 0.453 for the female. The same is true of the pancreas-adrenal weight association. The zero order coefficients of 0.367 ± 0.053 in the male and 0.491 ± 0.047 in the female are only reduced to 0.225 and 0.293 respectively, when body weight is held constant, and positive specific association is maintained when in addition the gonads, thyroid, hypophysis and thymus are collectively stabilized for. The 5th order value is 0.132 in the male and 0.238 in the female. Positive specific

weight association can be assumed to indicate specific functional association, at least in the quantitative sense.

The values just given show that there is a specific functional as well as weight association between adrenals and gonads, and adrenals and pancreas. The reciprocal relationships of the triad

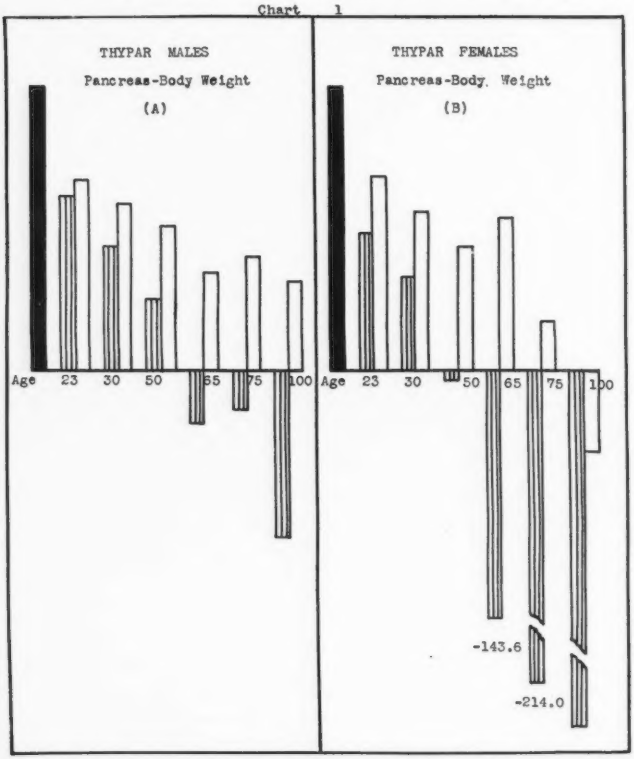

Chart 1. Comparing growth retardation of pancreas and body weight after thyroid removal from A—Males and B—Females at the stated ages. Flat black columns represent control growth (100 per cent); outline columns, growth of test animals in terms of that of controls. Order of sequence given by the superscript.

are represented in the accompanying diagram. This shows that any size relation between gonads and pancreas is conditioned by the adrenals.

Now it was shown in the preceding paper (4) that a pubertal and post-pubertal initiation of thyroid deficiency causes re-

gression of adrenal growth, and that this is largely dependent on the gonads. The evidence would, therefore, indicate that it is this adrenal regression which directly determines the increased sensitivity of the pancreas to thyroid deficiency.

The pancreas should be expected to be more closely dependent on the adrenals than on the gonads from the known fact of the functional interrelations in carbohydrate metabolism, and the fact that thyroid function is also concerned in this phase of organic activity through its participation in the regulation of the metabolic level.

From the foregoing the conclusion is justified that the pancreas shows no specific growth relation to thyroid activity.

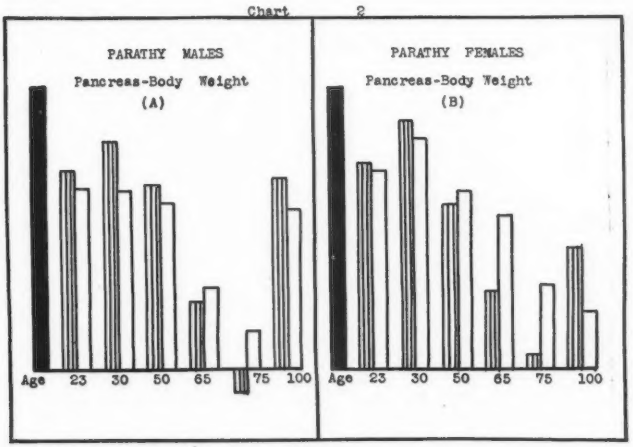

Chart 2. Comparing growth retardation of pancreas and body weight after parathyroid removal from A—Males and B—Females at the stated ages. Flat black columns represent control growth (100 per cent); outline columns, growth of test animals in terms of that of controls. Order of sequence given by the superscript.

A further contribution to the increased sensitivity of the pancreas in the 65, 75 and 100 day old series is the normal cessation of growth of the organ between 65 and 75 days of age (Table 1). The drop in normal growth capacity from the preceding period value of 2.9 in the male and 2.4 in the female is to —0.02 and 0.02 respectively. While the later periods show some slight growth this is very small in comparison to that of the prepubertal periods. The rôle of the normal growth capacity in

determining the degree of resistance to thyroid deficiency has been discussed in other papers and need not be repeated.

Now the pancreas is a digestive organ, a large proportion of the metabolic processes of which are concerned in its proper function rather than in the business of growth. It can be as-

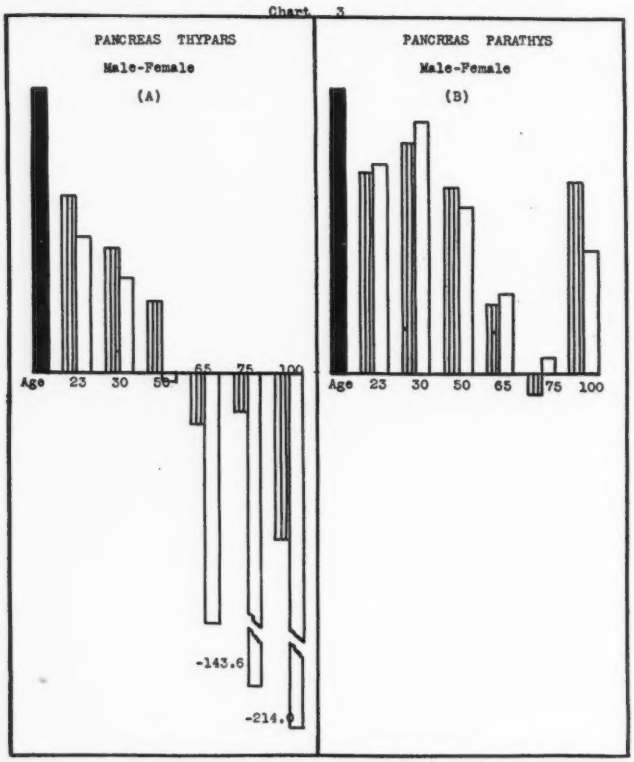

Chart 3. Sex comparison of pancreas response to A—Thyroid removal and B—Parathyroid removal at the stated ages. Flat black columns represent control growth (100 per cent); outline columns, growth of test animals in terms of that of controls. Order of sequence given by the superscript.

sumed that the ratio of the energy used in work done in functional activity and work done in growth is higher in the pancreas than in the body as a whole. This being so, it would be expected, on the basis of the deductions outlined elsewhere (2), that the pancreas would be more sensitive than the body as a

whole to the lowering of the metabolic level consequent on thyroid removal. It would also be expected that the difference would increase, if and when pancreas growth ceases while total function increases and body growth continues. That being consistent with expectation, the foregoing is adequate present interpretation of the pancreas being more sensitive than the body as a whole to thyroid deficiency.

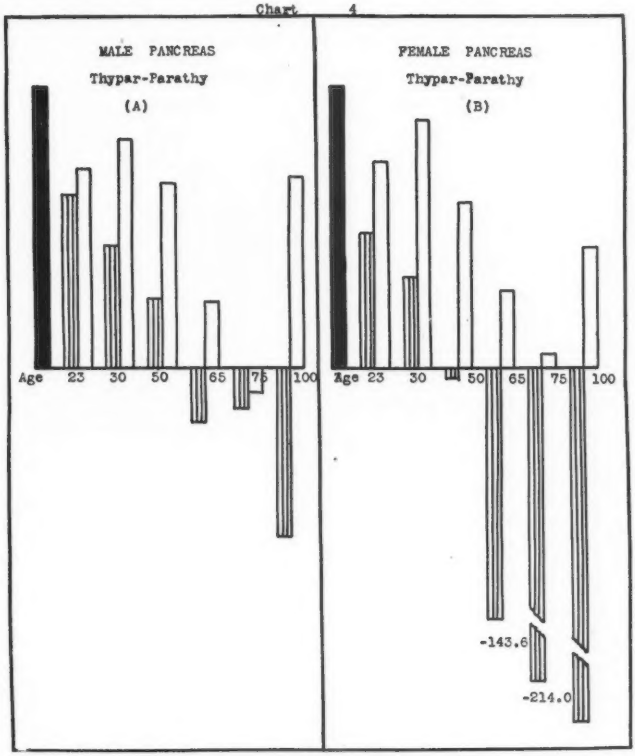

Chart 4. Comparing the effect of thyroid and parathyroid deficiency on pancreas growth in A—Males and B—Females. Flat black columns represent control growth (100 per cent); outline columns, growth of test animals in terms of that of controls. Order of sequence given by the superscript.

From Chart 3A it is seen that the pancreas of the female is more sensitive than that of the male. Since this sex difference increases markedly from puberty onwards it is probable that its

basis lies in large part in the fact that the gonad-adrenal weight association is greater in the female than in the sexually mature male. To this the lesser normal growth capacity of the female pancreas (Table 2) undoubtedly contributes. The discussion of this principle has been given in earlier papers.

Jackson (5) reports that dietary deficiencies in the young produce simple retardation of pancreas growth, while like defects in older animals yield an atrophy. The age differential responses to thyroid deficiency are similar in type. This fact is consistent with the assumption that the growth retardation which follows thyroid removal is fundamentally due to a condition of essential undernutrition (2).

THE INFLUENCE OF PARATHYROID DEFICIENCY

An inspection of Chart 2A and B shows that the growth retardation of the pancreas closely approximates that of the body weight in the age differences in response to parathyroid removal. The conclusion is that the growth processes of the pancreas are quite like those of the body as a whole in their relation to parathyroid deficiency and that they are therefore not specifically related to parathyroid activity.

The chart also shows that puberty conditions a change in the relative degree of retardation. It is doubtful if this is attributable to any specific gonad-pancreas relation. Rather it is to be taken as an expression of an individual organic reaction to the general physiological changes produced by adolescence. It is hardly an incretory matter since the direction of difference between pancreas and body weight, once puberty is over, becomes the same (in the 100 day old series) as in the pre-pubertal groups.

Chart 3 B shows that no consistent sex differences are present. A detailed analysis of those which do exist is hardly worth while, since interpretation is impossible.

Turning now to Chart 4A and B where the response of the pancreas to thyroid deficiency is compared with that to parathyroid deficiency, a marked difference in the reaction to the two types of glandular lack is noted.

The pancreas is much more sensitive to thyroid than to parathyroid deficiency. This general difference demands interpretation. I am inclined for the present to believe that its basis lies

in the difference between the two types of metabolic disturbance induced. Thyroid removal primarily determines a lowering of the metabolic level, a secondary effect of which is a lowering of the amounts of material available for growth. Parathyroid removal determines an increased utilization of metabolites and a lowering of the nutritional level (2). The two deficiencies are thus in last analysis provocative of essentially the same condition in so far as growth is concerned: i. e., a state of undernutrition. But the primary metabolic disturbance is in general opposite in character. Hence it is probable that the lesser growth retardation of the pancreas subsequent to parathyroid removal is an expression of a relatively lesser disturbance of the metabolic rate, to which the organ is peculiarly susceptible by virtue of its functional specificity, and also to a lesser diminution in the amounts of material available for growth.

A second matter for clarification is the marked divergence in character of the response to the two types of deficiency when initiated during and after puberty. The reaction to thyroid deficiency, based on direct adrenal interference initiated by gonad response and conditioned by a specific thyroid-adrenal association, is discussed in the preceding section. In the case of the parathyroidless animals the picture is obscure. There is correlation between adrenal and pancreas reaction up to the 100 days old series (4). Here the adrenals undergo atrophy, while the pancreas is only retarded. The adrenals diverge from the body weight response while pancreas growth is less retarded. While the pancreas-adrenal parallelism is only fair in the male, the removal of the parathyroids at 100 days of age produces the same differential reaction as in the female. In view of this fact, and the fact that the pancreas generally parallels the body weight changes with change in age at time of glandular removal, I am inclined to believe that while the adrenals may be a direct factor in the determination of the pancreas response to parathyroid removal, they are only a minor factor, save possibly during puberty. The major factor is the dependence of the pancreas on the effectiveness of the growth processes of the body as a whole.

On the other hand, as shown in an earlier paragraph, there is a positive specific weight, and hence functional, association between pancreas and adrenals in the adult rat. It should be

expected, therefore, that if this is a large factor in determining pancreas response to thyroid deficiency, it should also be evident as a factor in parathyroid deficiency initiated at 100 days. But such is not the case. It is, therefore, clear that a factor is lacking in the adult parathyroidless which is present in the thyroidless groups. The only factor which present evidence justifies us to believe is absent from the one and present in the other is a specific relation between the adrenals and the gland removed. There is a specific association between adrenals and thyroid. There is no proof of a specific relation between adrenals and parathyroid. The tentative conclusion is, therefore, justified that it is the presence of the thyroid-adrenal specific relation-

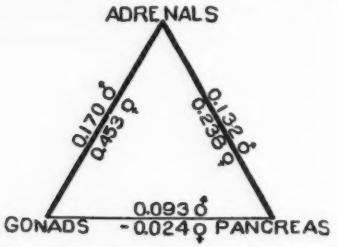

Diagram. Showing the adrenals as the pivotal point of gonad and pancreas relations. The figures are the values for the specific coefficients of correlation unconditioned by body weight, thyroid, hypophysis or thymus.

ship which determines the difference in type of pancreas reaction to thyroid and parathyroid removal at 100 days of age.

Regardless of the uncertainty of the details of relationship in the parathyroidless groups, a few significant facts are clear and these can serve as a starting point for further work leading to ultimate clarification.

SUMMARY AND CONCLUSIONS

A study of the pancreas response to thyroid and parathyroid deficiencies indicates clearly that the growth of the organ is not specifically related to the functional activity of either of these incretory glands.

In the thyroidless groups the dependence on the effectiveness of the growth processes of the body as a whole is the chief pre-pubertal factor determining the growth retardation. During and after puberty, when growth regression instead of simple retardation occurs, the gonads and the adrenal are participants

in determining the increased sensitivity. The connecting specific link is the adrenal.

In the parathyroidless groups the chief factor throughout is the dependence on the effectiveness of the growth processes of the body as a whole.

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THE OCCURRENCE AND PRESENT CHEMICAL STATUS OF THE FEMALE SEX HORMONE *

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The chemical identity of the female sex hormone has been established only between certain rather wide limits. The methods of assay are all biological and consist of the following:

1. Noting a growth producing action on the uterus, vagina and breasts of immature virgin rabbits, or of castrated rabbits (Figs. 1 and 2).

2. Noting the change produced by injection of extracts on the vaginal spreads obtained from spayed rats. The vaginal spread of the castrated rat consists mainly of leukocytes. When potent extracts are injected, after an interval of between twelve and forty-eight hours, the spread changes to squamous non-nucleated epithelium similar to the spread obtained from normal rats at œstrus (Figs. 3 and 4).

3. Noting the spontaneous, rhythmic contraction rate of the uterus of the white rat when suspended in a chamber filled with Locke's solution. The uterus of the castrated rat has a rhythm identical with that of the early interestrus period of the normal rat. The uterus of a rat whose vaginal smear had become positive by means of injection of potent extracts produces the slower contraction rate typical of estrus in the normal animal (21).

All of these methods are reliable and yield identical results. The method of assay based on vaginal smears, introduced by Allen and Doisy (1), is the most convenient because it does not involve the sacrifice of the animal and the results are available in a much shorter time. The introduction of this method of assay marked a very important advance in the technique for the

*Read before the Tenth Annual Meeting of the Association for the Study of Internal Secretions, Dallas, Texas, April 20, 1926.

study of the female sex hormone. All of the above methods of assay indicate that the hormone as obtained from the follicle liquid, the corpus luteum, placenta, and blood of animals in estrus is identical. Chemical studies to date lead to the same conclusion.

The occurrence of the female sex hormone in the follicle liquid was first reported by Frank (2). In the follicular liquid the hormone occurs in such quantities that no concentration is necessary, and the pure fluid itself will produce positive results by all methods of assay. Allen and Doisy (8), continuing the work of Frank, have shown that the hormone follows the lipoid fraction of the liquid and have further shown that the hormone can be freed from cholesterol by precipitating the cholesterol with digitonin. Studies on the hormone from the follicular liquid have given us no chemical information which we did not have from studies on the hormone obtained from the corpus luteum and placenta.

The occurrence of the female sex hormone in the corpus luteum has been established by many workers, among them Iscovesco (3), Hermann (4), Frankel and Fonda (5), Fellner (6), Frank and Rosenbloom (7), etc. These workers based their conclusions on the production of hyperplasia and hypertrophy of the uterus of the young immature virgin rabbit and of the castrated rabbit. With the introduction of the vaginal spread method of assay Allen and Doisy (1) report only negative results with their corpus luteum extracts. An obvious explanation is that they were unable to concentrate the active substance found in the crude lipoid extracts. These workers in a preliminary report assumed that the female sex hormone did not occur in the corpus luteum, at least of the hog.

In a second article (8) not only was the presence of the active principle in the corpus luteum denied but doubt was cast on all of the work of other investigators in this field who had reported the presence of the hormone in this body. The following is a quotation from that paper of Allen and Doisy:

"Although all of the authors referred to in the preceding paragraphs who used lipoid solvents may have obtained active extracts from whole ovaries or placenta, their findings of the hyperplastic substance in the corpora lutea casts doubt upon their observations. While it is not in our province to explain this confused situation, we may point out that in some cases it might be attributed to the use of

normal animals in testing for activity. Certainly none of the authors with the possible exception of Hermann has shown that this extract can restore to ovariectomized animals the full growth of the genital tract and the sexual manifestations characteristic of estrus."

This statement was based on the fact that Allen and Doisy were unable to obtain the vaginal spread reaction in spayed rats with their extracts of corpora lutea. Similar findings were more recently reported by Johnston and Gould (9) who failed to get a single positive reaction with 27 preparations of the yellow body. Our studies have shown that larger doses of corpus luteum extract must be given over a longer period of time than if follicle fluid be employed in order to secure hypertrophy of the uterus and mammary glands in either spayed or immature virgin rabbits.

Corpora lutea to the amount of 358 grams were obtained from 1800 grams of freshly slaughtered hogs' ovaries and, after being washed twice with water, were at once ground in a meat chopper and then with sand in an iron mortar. Five hundred fifty cc. of 95% alcohol was added. The mixture was thoroughly shaken and allowed to stand for 48 hours. The mixture was filtered and evaporated under reduced pressure to a watery sludge. This sludge was extracted twice with 50 cc. of benzene. The benzene extract on evaporation gave 2.7 grams of residue. A residue prepared in this manner is called "Stock B." This residue was suspended in olive oil making the concentration 25 mg./cc. An immature virgin rabbit injected with this material on alternate days for two weeks showed hypertrophy of the uterus and mammary glands and the result was indicated as plus 2.

Stock B was prepared from 545 grams of corpora lutea from 2200 grams of hogs' ovaries taken immediately after slaughter and tested in a similar fashion. The results with some animals, given only four injections, were indefinite. Animals, however, that were given seven injections over a period of two weeks showed a plus 2 reaction.

With the vaginal smear reaction results with the above material were negative with four injections of 25 mg. each and also with four injections of 50 mg. each. This led to intensive study of the preparation of corpus luteum extracts with the result that extracts have been obtained which now give all reac-

tions that extracts of follicle liquid and placenta will give, including the vaginal smear test demanded by Allen and Doisy.

In the preceding experiments the corpora lutea had represented a composite lot; no attempt had been made to classify them. It seemed possible that the hormone content of the corpus luteum might vary with the time that had elapsed since ovulation. Corpora lutea obtained from 4900 grams of ovaries from freshly slaughtered hogs were separated into classes corresponding to early vascularized, later vascularized, flourishing, and involuting; or, more simply, into bloody, bloody pink, pinkish yellow, and yellow.

Samples were taken and analyzed with the following results:

	Bloody	Bloody pink	Pinkish yellow	Yellow
Grams of each class	98	484	378	69
Moisture %	81	79.9	79.1	75.5
Crude lipoid (% of dry substance)	18.6	25.4	36.6	32.2
Nitrogen (% of dry substance)	12.6	10.68	10.45	10.38
Phosphorus (as % of pentoxide of dry substance)	2.68	3.38	3.44	2.42
Ash (% of dry substance)	4.63	5.26	4.92	3.95

Stock B was then prepared from each of these classes and suspended in olive oil with a dosage of 50 mg. per cc. Assayed by the vaginal smear test the results were as follows:

Class	Number of injections	Total dosage	Reactions 1st animal	2nd animal
Bloody	3	150 mg.	negative	negative
Bloody-pink	3	150 mg.	negative	negative
Pinkish-yellow	3	150 mg.	plus 1	plus 1
Yellow	3	150 mg.	plus 3	plus 2

The striking result is the concentration of the active material in the yellow class. Frank has offered the following explanation for these findings (10):

"At the time of follicle rupture a concentrated active extract, the follicle fluid, is poured into the peritoneal cavity and rapidly absorbed by the lymphatics. The granulosa of the follicle at that time has no connection with the blood stream; hence the product is stored. But immediately after ovulation, capillaries penetrate the ruptured follicle, and within a few hours the process of vasculariza-

tion places each secreting cell in intimate contact with one or more capillaries. From that time on the follicle, or better, its successor, the corpus luteum, is no longer a storage gland and the secretion is carried off as soon as formed. When full maturation has been reached or just passed, the blood channels begin to diminish before the secreting cells lose their activity. Therefore, for a short period a sudden transitory increase in the stored active substance is noted."

Concentration of the hormone by means of removal of fats, phospholipins and cholesterol was then attempted with the following results:

Class	Amt. of of crude material injected	Result on spayed rats	Amt. of purified material injected	Result on spayed rats
Bloody	150 mg.	neg. 48 hrs.	75 mg.	plus 3, 36 hrs.*
Bloody pink.	150 mg.	plus 2, 48 hrs.	75 mg.	plus 4, 36 hrs.
Pink yellow .	150 mg.	plus 3, 48 hrs.	75 mg.	plus 4, 18 hrs.
Yellow	150 mg.	plus 4, 36 hrs.	75 mg.	plus 4, 18 hrs.

More highly concentrated and purified material gave positive results with a total dosage of 45 mg. and 28 mg.

These experiments prove conclusively that the hormone is found in the lipoid fraction of the corpus luteum and that it is more concentrated in the yellow type of corpora lutea. Purified extracts produce hypertrophy of the uterus, vagina and breasts in immature virgin rabbits and in castrated rabbits. They produce the vaginal smear reaction in rats and the uteri of these spayed rats show typical spontaneous contractions in oxygenated Ringer's solution that are characteristic of oestrus. The material was fresh, obtained at the slaughter house and at once worked up. Therefore the possible but improbable objection of post mortem diffusion of follicle fluid into the corpus luteum cannot apply.

In a recent paper Allen and Doisy (11) apparently reverse, in part at least, their position as to the presence of the hormone in the corpus luteum. They state that the corpus luteum in women continues to secrete the hormone for a considerable period of time, even stating that the corpus luteum of the first and third months of pregnancy continues to secrete the hormone. The explanation is offered that the corpus luteum of the primate occupies a different position from that of the rest of the mammals. This is an extraordinary assumption since

*Plus 2 signifies squamous and nucleated epithelium, no leucocytes present.

the corpus luteum is actually found among monotremes in which the uterine portion of gestation is very short.

While our work proves conclusively by chemical methods that the hormone is found in the corpus luteum of the hog, this conclusion has been corroborated from a biological standpoint by Zondek and Ascheim (12). These workers implanted small portions of the tissue to be tested in the thigh of spayed mice and studied the effect on the vaginal spreads. Using human material they found no hormone in germinal epithelium, stroma

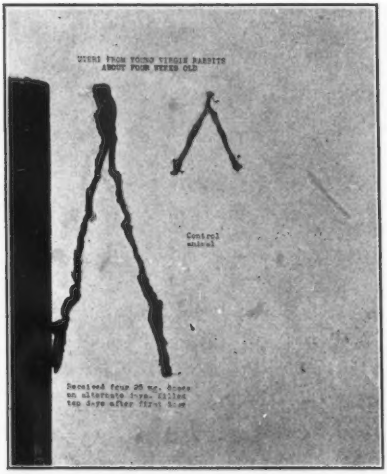


Fig. 1. Uteri from young virgin rabbits about four weeks old.

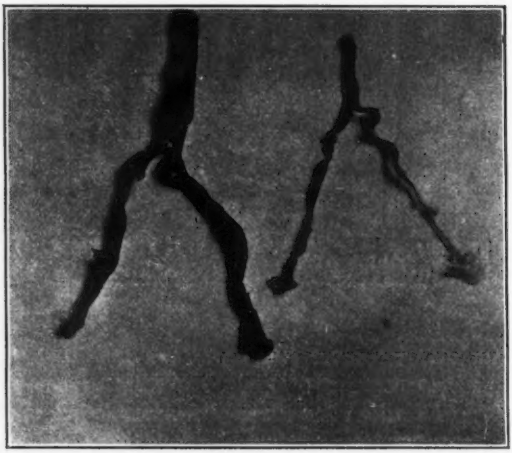
or primordial follicles. All larger follicles contained the female sex hormone. Implantation of the wall of ripening follicles also gave a positive result. Implantation of the flourishing corpus luteum always gave a positive reaction. Tests with liver and other tissues gave negative results.

These concordant results obtained by many investigators by different methods should end this long drawn-out controversy. Positive results based on material obtained with all precautions, assayed by all methods known, and repeated without failures after proper extraction, technic, and dosage had been worked out, must surely outweigh negative reports. The fail-

ures are ascribable to lack of concentration and subthreshold dosage.

As our researches and those of other workers indicate that the hormone found in the follicles, corpora lutea, and placenta are identical, we have used placentae as the base of our further work, because placentae are easily obtainable and contain the hormone in large quantity and are readily extracted. The chemical study of this, as of other hormones, is still far from complete. The question of chemistry resolves itself into two problems:

1. Is the physiologic activity of crude extracts due to the specific substances alone, or can these responses be brought about by other substances?



A B

Fig. 2. (a) Uterus of castrate rabbit injected with hormone from placentae. (b) Uterus of castrate rabbit not injected.

2. What is the chemical nature of these specific substances?

Zondek and Robinson (13) have reported the growth of the uterus and vagina of guinea pigs, in response to injections of aolan, histamin, tenosin and epinephrin. Our work, in which immature virgin rabbits were used, gave negative results with these substances. Negative smears were also obtained when we used spayed rats. Schroeder and Goerbig report positive results with liver lipoids (14). We obtain consistently negative

results with liver lipoid. Similar negative results were obtained by Zondek and Ascheim (12) with their implantation technic. The spontaneously ovulating guinea pig is an unsuitable test animal. It was mainly employed by the authors who reported positive results with non-specific substances. In addition the following control materials gave uniformly negative results:

Material	Test used*	Animal	Amt. injected
Olive oil	h. u.	rabbit	4 cc.
Ole-one	h. u.	rabbit	200 mg.
Ole-one	h. u.	rabbit	200 mg.
Liver	h. u.	rabbit	200 mg.
Butter	h. u.	rabbit	200 mg.
Histamine	h. u.	rabbit	6 mg.
Tenosin	h. u.	rabbit	
Aolan	h. u.	rabbit	
Adrenaline	h. u.	rabbit	
Histamine	v. s.†	rat	
Tenosin	v. s.	rat	
Aolan	v. s.	rat	
Adrenaline	v. s.	rat	
Brain	h. u.	rat	100 mg.
Brain	v. s.	rat	100 mg.
Liver	v. s.	rat	75 mg.
Liver	v. s.	rat	82 mg.
Testes	h. u.	rabbit	100 mg.
Testes	v. s.	rat	100 mg.
Adrenal cortex	h. u.	rabbit	150 mg.
Adrenal cortex	v. s.	rat	150 mg.
Adrenal medulla	v. s.	rat	75 mg.
Thymus	v. s.	rat	100 mg.
Linseed oil	v. s.	rat	100 mg.

Assuming the physiologic response to be due to a specific substance, what are its physical and chemical properties? Iscovesco, the pioneer, showed that the lipoid fraction of the placenta contained the active principle (3). Similar results were obtained with the lipoids of the corpus luteum, placenta and follicle liquid by many other workers. Our work has confirmed the statement that the active principle is soluble in lipoid sol-

*h. u. indicates hypertrophy and hyperplasia of the uterus in young immature virgin rabbits.

†v. s. vaginal smear test on spayed rat.

vents. We also find it to be soluble in water under certain conditions, depending on the presence of other extractives and on methods of purification. Lowe and Laquer report a water soluble preparation of which 1 mg. gave a positive reaction in the spayed mouse (between 1/20 and 1/8 of the rat dose).

In 1915, Hermann reported the isolation of an active preparation from deblooded placenta free from nitrogen and phos-

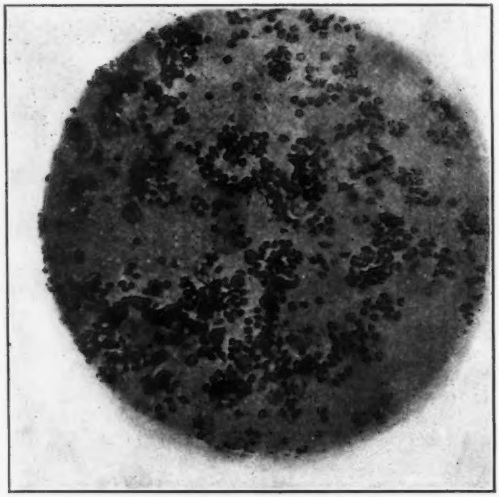


Fig. 3. Cell types found in smear made from castrate rat. Leucocytes.

phorus (4). Frankel and Fonda confirmed this work (5). The most active extracts of Giesy (15) were free from nitrogen and phosphorus. Our work is in harmony with these findings. The preparations of Allen and Doisy contain about 1% nitrogen.

The active substance shows a very high degree of thermostability. All workers are agreed on this property. Herrmann (4), Frankel and Fonda (5), and Frank and Lee have attempted purification by distillation under high vacuum at temperatures ranging from 190° C. to 250° C. without loss of activity. A purified fraction distilling below 100° C. at 30 mm. recently gave us positive results.

The active principle can be extracted from alkaline solutions with ether. This would seem to eliminate acid groups.

It can also be extracted from acid solutions with ether which would seem to eliminate basic groups. Esters seem to be eliminated on the following grounds. Giesy found that the activity was not destroyed by digestion with soy bean lipase. In our work we have saponified the active lipoid fraction with half normal potassium hydroxide, sodium methylate, sodium butylate and sodium ethylate for periods of 72 hours without loss of activity. When the saponified mixtures are evaporated to dryness and extracted with ether, we find that these extracts are physiologically active. In other words, it is either a non-saponifiable material, or, if it is an ester, the activity resides in the alcohol of high molecular weight liberated by the saponification process.

TABLE V

Grams of Stock B.....	.4892	.4874	.4952	.6750	.6656
Cc. 0.5N KOH used.....	50	50	50	50	50
Time of saponification (hrs.)	24	36	48	60	72
Weight of ether extract of dried residue	.2780	.2660	.1938	.2780	.2390
Results of assay — vaginal smear test	plus 4	plus 4	plus 4	plus 4	plus 4
Mg. injected	37.5	37.5	37.5	37.5	37.5
Weight of ether extract from dried residue upon acidifying after alkaline extraction	.1358	.1844	.2390	.3280	.4050
Assay of ether extract of acid material	neg.	neg.	neg.	neg.	neg.

The results show that the active principle can withstand saponification with alkali for a long period of time without loss of activity. These results are in disagreement with those of Dickens, Dodd and Wright (16), who state that the hormone is slowly destroyed by saponification for 40 minutes in the presence of hydrogen.

Frankel and Fonda (5) report the isolation of the active principle which they describe as thick, viscous light yellow oil with turpentine-like odor, insoluble in water but soluble in all lipoid solvents. They ascribe the formula $C_{32}H_{52}O_2$. One atom of oxygen, according to their work, is present in a hydroxyl

group and the other in a carbonyl group. The compound is unsaturated and contains two double bonds. Several derivatives, esters, phenylhydrazone, oxime and tetrabromid were obtained by these authors to substantiate their suggested formula. The pure substance shows cholesterol reactions which Frankel and Fonda take to indicate a relationship to cholesterol and bile acids.

Frank (17) and Doisy, Ralls, Allen and Johnston (18) found that the active principle can be freed from cholesterol and thus a product is obtained which does not give cholesterol reactions. This work indicates that the substance isolated by Frankel and Fonda was at least contaminated with cholesterol. The derivatives of the "pure substance" obtained by them were not

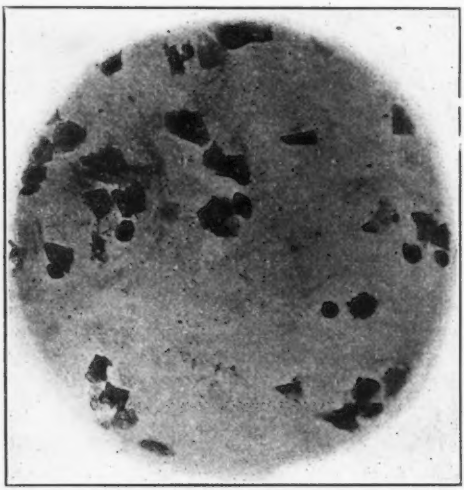


Fig. 4. Cell types found in smear made from castrate rat injected with active hormone preparation from placentae. Squamous epithelial cells.

reconverted into the original compound to see whether the reisolated substance was active. This crucial test would determine whether the derivatives were from impurities or from the pure substance. Work in our laboratories does not substantiate the statement of Frankel and Fonda that the active principle forms a hydrazone.

We have obtained a phenylhydrazone from the lipoid fraction of placenta. The mother liquor after removal of the phenyl-

hydrazone and excess phenylhydrazine is still active physiologically. This might be due to incomplete precipitation. When the phenylhydrazone is decomposed by being allowed to react with glucose, a lipoid is liberated which is inactive physiologically.

The active principle is an unsaturated body absorbing bromine from carbon tetracloride solution. It is oxidized by exposure to air to a resinous mass darker than the original substance. This oxidation is accelerated by light. The oxidized product is inactive physiologically. This indicates that the active principle is an unsaturated body and not merely associated with unsaturated bodies.

A number of experiments were carried out to test the effect of PCl_5 and PCl_3 on the hormone. The results are shown in table VI.

TABLE VI

Amount of stock B	Reagent	Amount of reagent	Temperature °C.	Time of heating	Amount injected	Smear reaction
1.056	PCl_5	1.1 g.	145	2 hrs.	100 mg.	neg.
1.15	PCl_5	1.2 g.	110	2 hrs.	75 mg.	neg.
1.10	PCl_5	1.1 g.	110	1 hr.	75 mg.	neg.
1.07	PCl_3	1.2 g.	75	1 hr.	37.5 mg.	plus 4
1.00	PCl_3	1.1 g.	70	2 hrs.	37.5 mg.	plus 4
1.14	PCl_3	1.2 g.	70	2 hrs.	37.5 mg.	plus 4

The destruction by PCl_5 is apparently by oxidation. The conclusion is confirmed by the fact that after PCl_5 treatment the lipoid shows a large decrease in the Iodine number, i. e., from 150 to 74. The fact that the active substance is not destroyed by PCl_3 would indicate that either the hormone does not contain the hydroxyl group or replacement of the OH by Cl has no effect on activity. Halogens are found in our lipoid preparations only after PCl_3 or PCl_5 treatment.

Today we are also able to show that the female sex hormone is present at times in demonstrable quantity in the circulating blood (19). Blood has been collected from hogs in estrus and anestrus. Following the methods developed in connection with the work on placenta and corpus luteum, lipoids have been obtained which, on injection into spayed rats, give the usual vaginal smear reaction. This phase of the problem is still under investigation. Loewe (20) reports similar results.

From the studies up to this time we can say that the hormone may be found in the follicular liquid, corpus luteum, pla-

centa, and blood of females in estrus. The term follicular hormone or ovarian hormone is therefore not sufficiently broad. "Female sex hormone" covers the source of origin as well as the biological function of the substance in question and should therefore be employed. As the hormone can be found in the follicle, corpus luteum and placenta, and as the corpus luteum succeeds the follicle, and the placenta the corpus luteum, the term "gestational gland," as a suitable appellation for this triad, has been suggested.

The active substance is a thermostable lipoid of high molecular weight. It is soluble in all lipoid solvents and also in water under certain conditions. It contains only carbon, hydrogen and oxygen, and possibly merely carbon and hydrogen. It is not an ester or a fatty acid. It does not give cholesterol reactions. It does not contain an active carbonyl group. If it contains the hydroxyl group this group is not essential to its activity. It is an unsaturated compound and is inactivated by attacking the double or triple bonds.

The female sex hormone is a specific substance elaborated by the gestational gland, taken up by the lymph and blood stream and selectively utilized only by Mueller's tract and the breasts.

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SOME CHEMICAL AND PHYSIOLOGICAL PROPERTIES OF THE HORMONE OF THE LIQUOR FOLLICULI.*

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INTRODUCTION

Since the title of this paper may give rise to the question of whether or not the active principle of the placenta and corpus luteum differs from that of the liquor folliculi, we may state at the outset that such physiological data as are available and such chemical data as are admissible for deciding the question indicate the probable identity of the hormone from the three sources. Due to the failure as yet of chemical isolation of the hormone from any source and to the fact that it seems better to us to refrain from a name until a logical one based on its chemical constitution can be given, and since all of the observations described in this paper were made upon liquor folliculi from hog ovaries, we prefer to refer to the product with which we are working as the hormone of the liquor folliculi.

Within the past few months a number of papers upon the ovarian hormone obtained from liquor folliculi have appeared. Most of them appear to be of a preliminary character and therefore cannot be discussed very intelligently. Zondek and Brahm (1925) and Laqueur, Hart, deJongh and Wijsenbeek (1926) agree that the hormone may be obtained in the aqueous filtrate from the deproteinized follicle fluid. This filtrate may be concentrated in vacuo, yielding finally a potent aqueous solution of the hormone.

Laqueur et al. in studying the chemical nature of the solids of their extract have concluded that protein, cholesterol, amino

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nitrogen, phosphorus, and the hydroxyl group are absent. Some of the extracts obtained contained one mouse unit per 0.01 mg. or even less of solids. Inasmuch as the use of different test animals renders comparison of potency difficult, we cannot definitely assert that this is the most active product thus far reported in the literature, but at any rate this statement is probably correct.

Both Laqueur et al. and Dickens, Dodds and Wright have failed to obtain yields (in physiological units) as large as those reported in our earlier paper (Doisy, Ralls, Allen and Johnston, 1924). Two of us (J. O. R. and E. A. D.), working independently, have made many preparations in which from 500 to 1400 rat units per liter of liquor folliculi were obtained. We suspect that both groups of investigators may have tried to improve our published procedure with the object of obtaining a purer product and in so doing lost some of the efficiency of the extraction process and therefore obtained smaller yields.

Upon still another point the conclusions of Dickens and Dodds disagree with ours, in that they report finding very little more hormone in the liquor folliculi than in an equal weight of ovaries. In this case the findings of Laqueur et al. are in accord with ours. Though we have made only a few preparations directly from whole ovaries during the past two years, we have found nothing to cause us to alter our earlier conclusion that the liquor folliculi of mature follicles contains much more of the hormone than other ovarian structures.

PREPARATION FROM LIQUOR FOLLICULI*

Liquor folliculi collected by aspiration from fresh hog ovaries is preserved with two volumes of 95 per cent alcohol until one liter is obtained. The hormone seems to retain its potency unimpaired under these conditions for as long as sixteen months. The average yield from a large number of preparations is about 850 units per liter of liquor folliculi; from two recent batches, one of which had been kept for six and the other sixteen months, we obtained 1390 and 1200 units respectively.

*See article by Ralls, Jordan and Doisy in the Journal of Biological Chemistry of August, 1926.

The proteins which are coagulated by the preservation in alcohol are filtered off and extracted in a Soxhlet apparatus; meanwhile the filtrate is being distilled to dryness in vacuo. After completion of the extraction, the extract is distilled from the same flask that was used for the filtrate. The dry residue is taken up in about 150 cc. water, any free fatty acids neutralized with sodium hydroxide, and the aqueous solution extracted four times with ether in a separatory funnel. Study of the individual extracts showed that about 80 per cent of the hormone was removed by the first ether extraction and that only 2 or 3 per cent remained in the aqueous phase after the fourth extraction. The ether extracts are combined, washed with dilute alkali and water and then distilled. The dry residue is taken up in 100 cc. 70 per cent alcohol and the cholesterol removed by five extractions of the alcoholic solution with 25 cc. of petroleum ether. This extraction is important for several reasons: (1) The hormone is soluble in the alcohol and only the merest traces pass into the petroleum ether. (2) The reverse is true for cholesterol. (3) Other solids which are present as impurities pass almost completely into the petroleum ether. This advantageous purification removes practically all of the cholesterol and about 95 per cent of the solids, leaving virtually all of the hormone in the alcoholic solution. The alcoholic solution is then distilled to dryness in vacuo and the residue taken up in ethyl ether. A small amount of residue which is insoluble in ether may be discarded as it contains little or none of the hormone.

Possibly one or two points in the above procedure deserve comment. We are not at all certain that of all of the possible combinations of solvents those chosen are the most satisfactory, but the method outlined gives a much better product with considerably less work than could be attained by our earlier technique. It is possible that some other concentration than 70 per cent alcohol might lead to better results; this point is under investigation and preliminary data indicate that a product at least four times as potent can be obtained by reducing the percentage of alcohol. One milligram of the preparation thus obtained equals 100-150 rat units, whereas 1 mg. of the solids obtained by using 70 per cent alcohol contains only 25-35 units.

CHEMICAL REACTIONS OF THE HORMONE OF THE 0.04 MG. PER RAT UNIT PRODUCT

The heading of this section requires some explanation. We prefer to state conservatively the effect apparently produced by certain procedures upon a product of the potency of 0.04 mg. per rat unit. From certain of our observations, it seems probable that fully 75 per cent and possibly much more of this preparation is not hormone. It is conceivable and indeed probable that certain reactions given by this product may not be given by the pure hormone, hence our caution in the interpretation of results.

Hydrolysis:

(a) Sodium ethylate. No effect upon potency.

(b) Lipases.

Pancreatic. No effect upon potency.

Liver. No effect upon potency.

Castor Bean. No effect upon potency.

Hydrogenation. No effect upon potency, visible decolorization.

Bromine. Complete loss of potency.

$KMnO_4$. Complete loss of potency.

Acetyl chloride. Marked loss of potency.

Benzoyl chloride. Moderate loss of potency.

Naphthylisocyanate. Moderate loss of potency.

Phenylhydrazine. No loss of potency.

Dry heat at 179° at 0.1 mm. for 2⅓ hours. Over 60% loss of potency.*

No attempt was made to isolate any of the products which may have been formed, but the reaction was carried out with minimal technical loss, the excess reagent removed and the residue injected into spayed rats.

Ultimate Analysis: In the chemical work our available supply of the hormone was so limited that we have had recourse to Pregl's (1923) system of microanalysis; when his methods were not suitable, we have modified them or devised new ones to fit the occasion. The results of our analytical work are given below:

*Attention should be called to the chemical investigations of Herrmann (1915), Fraenkel and Fonda (1923) and Giesy (1920). Most of the above reactions have been used in their studies of the hormone obtained from the placenta and the corpus luteum; our report is added as a characterization of a product of definite purity made by a clearly stated procedure from hog liquor folliculi.

Carbon, 80.8%; Hydrogen, 10.5%; Nitrogen, 0.93%; Phosphorus, 0.0%; Molecular weight, 458.

PROPOSED REQUIREMENT OF A PREPARATION FOR CLINICAL USE

Though we feel certain that the hormone under discussion may be of therapeutic importance, our clinical work has not progressed sufficiently to warrant any conclusions. Organotherapy at present in the ovarian field has both its strong proponents and its vigorous opponents. Both Frank (1922) and Novak (1922) have recently stated that they believed the present day commercial preparations lacked efficacy. We can corroborate their statements with respect to the hormone which produces growth and hyperemia of the genitals. Of a large number of preparations of both foreign and domestic manufacture tested, we have not found one which contained 1 rat unit in three ampoules or in several doses to be taken per os. Of the foreign investigators Zondek (1925) has called attention to the necessity of biological assay and has used the smear method proposed by Allen and Doisy (1923) on mice. He has reported that two of the products tested are potent, and Hannes (1925) using those preparations reports favorable results.

Tentatively we suggest the following requirements for an extract for experimental clinical work: (1) that the hormone be dissolved in an innocuous aqueous medium; (2) that it be assayed by the smear method; (3) that it be ampouled to contain 5-10 rat units per 1 cc.; (4) that ample tests of its sterility and keeping qualities be made; (5) that it should have no effect upon the blood pressure of anesthetized dogs. Taking up these points in the above order:

1. Though we have used "mazola" (corn oil) as the solvent for injection during routine testing, we cannot advocate its injection into patients. Mazola and probably other oils injected subcutaneously produce hard lumps which may slowly disappear or may produce granulomata. We are frequently forced to discard rats which have been used for a number of tests as there is scarcely any good space left for injections. Interesting results upon the local picture produced by the subcutaneous introduction of mazola alone and mazola containing the hormone have been reported by Burrows and Johnston (1925).

From the foregoing it is easily understandable why we prefer aqueous solutions or emulsions for clinical use. As many investigators have stated that the hormone is not soluble in water, it may be necessary to find some agent which will permit the formation of stable colloidal solutions. We tentatively suggest from some preliminary work that a dilute solution of sodium soaps of unsaturated fatty acids in 0.9 percent NaCl might be a satisfactory vehicle for the suspension of the hormone. No ill effects were observable in rats after the subcutaneous injection of such solutions of the hormone.

2. The vaginal smear method of the testing of ovarian extracts (Allen and Doisy, 1923) has proved to be convenient, economical and accurate. Tests of potency can be made in 48 hours on either castrated female rats or mice. Each animal may be used a number of times extending over a period of two or three months.

The accuracy of standardization of extracts will naturally depend upon the observance of a number of conditions and the use of a sufficient number of animals to minimize the errors of biological reactions. Among the factors which may affect the response of the test animal, the following should be mentioned: age, weight, diet, interval between oophorectomy and first artificial oestrous, and interval between artificial oestrous reactions.

In an earlier paper we defined (Doisy, Ralls, Allen and Johnston, 1924) the rat unit as the minimal amount of hormone injected subcutaneously in three equal portions at four hour intervals, which would produce oestrous in a spayed rat weighing from 140-160 gms. It seems that animals of greater weight will require a larger amount of hormone. Loewe and Laqueur, who have used the mouse instead of the rat, report that the mouse unit is from $\frac{1}{8}$ to $\frac{1}{4}$ of the rat unit, which is about the ratio of the weight of these species.

As might be expected, pronounced atrophy of the uterus due to lack of hormonal stimulus renders the animals less sensitive, and therefore the results less accurate. We find the most consistent results are obtained by injecting our animals about every sixth day. In case three or four preparations in succession are negative, we find it is desirable to "prime" the rats by

injecting an aqueous solution of hormone which will certainly give an oestrous smear.

Though the enumeration and execution of these various considerations may seem burdensome, the results seem to justify the precautions taken. Loewe has reported that the mouse smear method has an accuracy of 20 percent, and our impression gained from our work is that this figure is approximately correct.

Table 1

Effect of Sterilization with 15 lbs. Steam for 15 minutes on Aqueous Solutions of the Hormone.

No.	Before Autoclaving		After Autoclaving	
	Vol. Injected	Test	Vol. Injected	Test
6b11	0.90 cc.	—	1.05 cc.	+
	1.20 "	+	1.20 "	+
G20492	0.07 "	—	0.10 "	—
	0.10 "	+	0.15 "	+
31379	0.15 "	+	0.10 "	±
	0.20 "	+	0.12 "	±

SYMBOLS: + more than 1 rat unit in volume injected.
± not quite a rat unit in volume injected.
— less than 1 rat unit in volume injected.

3. The activity of the preparation to be used clinically is a matter for experiment. Based upon the ratio of the weight of a test rat and a 50 kilogram woman approximately 400 rat units would be necessary to produce oestrous in the latter, if that were possible. We suggest that for experimental clinical work a solution containing 10 rat units per 1 cc. be used. Since it seems from our work with animals that excessive dosage produces no ill effects, any danger to the patient seems very remote. As many as 20 rat units have been injected into a rat without detectable harmful results.

4. Ample tests of the bacteriological sterility are of such well-known necessity that their importance may be dismissed without further comment. The method of sterilization employed may depend upon the stability of the hormone. Mazola solutions may be autoclaved at 15 lbs. for 15 minutes without appreciable loss of potency. Our preliminary data of Table I upon aqueous solutions so sterilized indicate that possibly a slight loss of potency occurs. Autoclaving seems so much easier and safer that we prefer to use this procedure, providing subsequent work

shows that our preliminary observations are correct. In the event that future work shows that the hormone cannot withstand the sterilization with steam under pressure, the Berkefeld filtrate may be used.

During the experimental stages at least, repeated tests should be made to assure oneself of the potency of the solution being used. For our own experimental clinical work we propose to secure a large stock of ampoules filled from the same batch

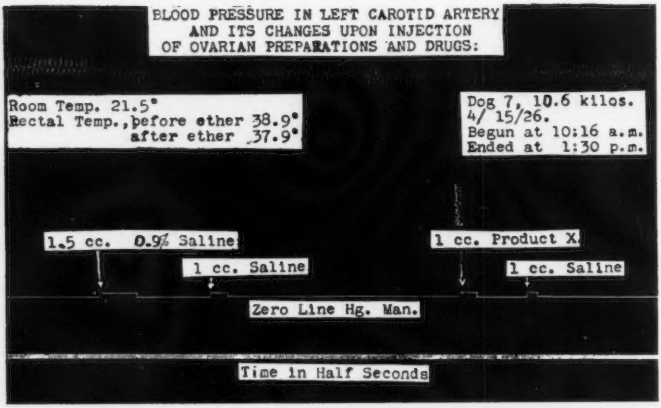

Fig. 1a. Blood pressure in left carotid artery and its changes upon injection of ovarian preparations and drugs.

of material. At least twice per week an ampoule will be tested for potency. By using this procedure failure to attain clinical results cannot be ascribed to a possible unknown deterioration of the extract.

5. *Effect of Injections of the Hormone upon Blood Pressure.* As tissue extracts frequently produce changes of blood pressure, it was deemed advisable to determine whether purified preparations of the hormone had any such effect. Of course, we still do not have the pure hormone, so that we cannot interpret our results with certainty, but one of our preparations has been purified to such an extent that we feel that a report of its effect upon blood pressure is worth while.

Cannulas were inserted in the carotid artery and femoral vein of dogs under ether anaesthesia. The blood pressure was recorded with a mercury manometer whose connecting tubing was filled with

a 5 per cent solution of sodium citrate. Time was recorded in half seconds and a signal magnet used for recording the time of injections also traced the zero line of pressure. The solutions tested were injected into a short rubber tube attached to a cannula in the femoral vein and then washed out of the cannula with 1 cc. of normal saline solution. The solutions tested were: normal saline; an aqueous suspension of 10f which was one of our purified preparations; an aqueous solution of one of our crude products; an aqueous suspension, product x, prepared by a pharmaceutical house; and an olive oil solution, product y, prepared by another company.

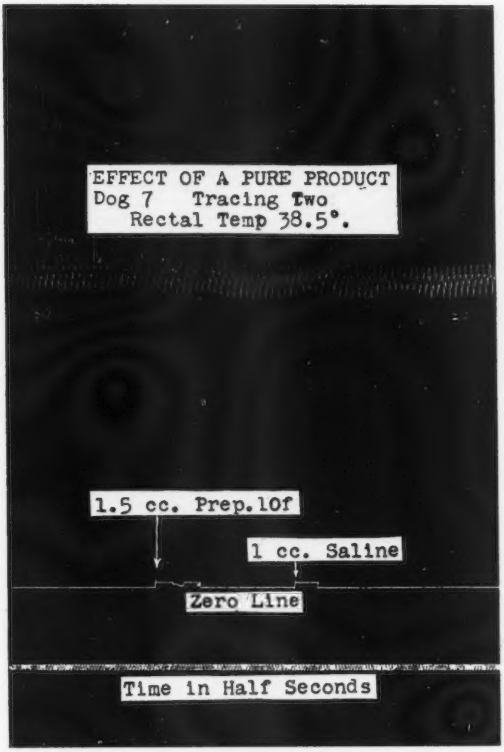

Fig. 1b. Effect of pure product.

Comparing the mean blood pressure and the heart rate (counted for 10 sec. periods) of Fig. 1a it may be seen that the pressure was 126 mm. and the rate 168. After the injection of 1.5 cc. and 1cc. of 0.9 percent saline, the values were 126 mm. and 180; and following the introduction of 1 cc. of product x,

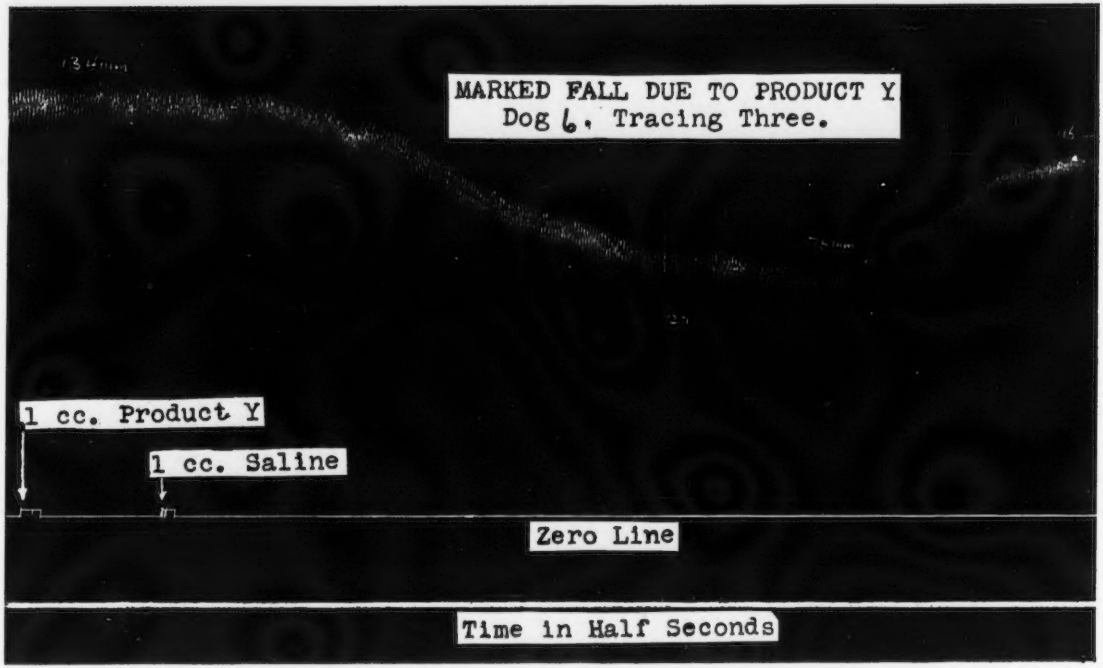

Fig. 1c. Marked fall due to product Y.

which contained 10 rat units, the blood pressure was 126 and the rate 174 per minute.

Figure 1b is a record of the effect of injection of 1.5 cc. of 10f containing 6 rat units. Blood pressure: before, 127 mm.; after, 127 mm. Heart rate: before, 180; after, 174.

Figure 1c shows the result of injecting 1 cc. of product y

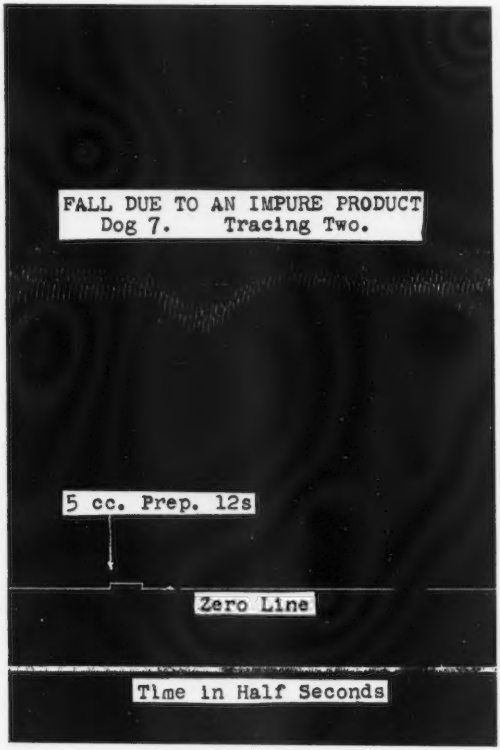

Fig. 1d. Fall due to an impure product.

which contained 5 rat units. The blood pressure at the time of injection was 134 mm. and the rate 156; after 4 minutes, during which a slow fall of blood pressure occurred, these values were 76 and 162 respectively. Shortly thereafter the blood pressure began to rise, and within 2 minutes had risen to 116 mm.

Figure 1d is a graphic record of the effect produced by the injection of 5 cc. of a crude solution of the hormone prepared from liquor folliculi. The mean blood pressure fell rapidly from 126 mm. to 110 mm., but recovered promptly.

Our results upon the effect of purified preparations of the hormone agree with those obtained by Dr. C. M. Gruber working with Allen and Doisy during the summer of 1923, in which only preliminary tests of relatively crude products were made, and with those recently reported by Laqueur and his coworkers (1926) using their own purified product made by a different method. It seems probable then that the hormone has no effect upon blood pressure and that preparations should therefore be tested upon the blood pressure before being used for experimental clinical work.

SUMMARY

1. A simple method of preparing potent preparations of hormone from liquor folliculi is briefly described.

2. Some of the chemical and physiological properties of this hormone are discussed.

3. The requirements that a preparation for experimental clinical work should meet are suggested.

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STUDIES ON VIGOR. VIII. THE EFFECT OF SUBCUTANEOUS INJECTION OF CORPUS LUTEUM EXTRACT ON VOLUNTARY ACTIVITY IN THE FEMALE ALBINO RAT.

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Although the sterilizing effect of corpus luteum extracts has been shown by Haberlandt (1), Knaus (2), and Papanicolaou (3), how the active principle produces its effect has not been made clear. The correspondence between the sex cycle and voluntary activity as described by Wang (4), and Slonaker (5) in the white rat suggest the possibility of throwing some light on the question by observing what effect such extracts exert upon this activity. Presumably any positive effect observed would be ascribable to an influence of the corpus luteum upon the ovarian follicular development.

For this experiment thirty-eight rats were placed in recording activity cages (6) for "typing" when about seven months old. Within a few days they began to manifest the usual oestrual variation in activity, becoming much more active every fourth or fifth day. Fig. 1. After typing, fifteen were selected for the corpus luteum treatment and nine generally from corresponding litters for controls, the former group being somewhat the more active.

An aqueous extract of corpus luteum, excepting on one day, that prepared by Hynson, Westcott and Dunning, was then injected subcutaneously on alternate days in the amount of 0.2 cc. to each rat over a period of twenty days. The next two injections were of 0.4 cc. each, then three of 0.3 cc. each and for the remainder of the experiment 0.2 cc. each time. The treatment was discontinued after 37 days. During the last twelve days daily microscopic examinations of the vaginal secretion were made as a check on the wheel records. These revealed the same incidence between "heat" and high activity in the treated animals as in the controls. Although the graph

has in the forty days of preliminary recording a few secondary peaks, simultaneous "smear" and activity records made on a considerable number of animals in this laboratory show that these are not all due to estrus.

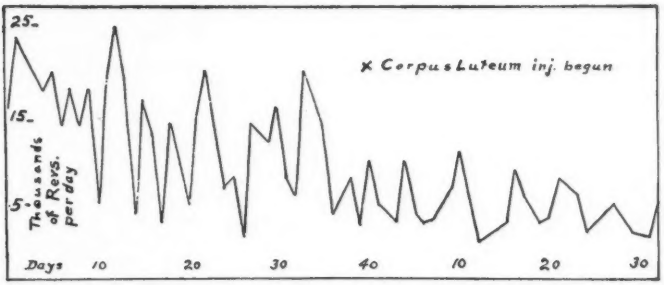

Fig. 1. Graph of cyclic activity of female white rat. The ordinates represent thousands of revolutions per day. The character x indicates when corpus luteum treatment began.

The results of the experiment are considered under two heads, periodicity of activity and average daily activity. Fig. 1 is a graph of the daily activity of an animal fairly typical as to both features. As will be readly seen, the oestrual variation in activity during the treatment with luteal extract does not significantly differ from that previous to treatment. This fact is also shown in table 1, which gives the average length of estrus cycle in each rat before and during the treatment, the averages

TABLE 1

Rat No.	A	B	Rat No.	A	B	Rat No.	A	B
1164	4.1	4.0	1095	4.5	4.8	1167	4.0	4.0
1077	4.4	4.3	1096	4.0	5.4	3268	4.1	4.6
1125	5.7	5.4	1100	5.0	4.0	3269	5.2	4.7
1091	4.1	4.7	1196	5.1	4.9	1110	4.8	6.0
1093	5.0	4.7	1128	4.1	4.0	1181	4.3	4.4

Table 1, showing average length of estrous cycle for each rat for 70 days before (column A) and 37 days during (column B) the treatment with corpus luteum extract. The average for the entire group of fifteen was 4.5 days before and 4.6 days during treatment.

for the entire group being 4.5 and 4.6 days, respectively. Composite graphs of each entire group were made by calculating the average daily activity over ten day periods. Table 2 gives these

averages for each animal and for each group, the latter being represented by Fig. 2. It will be seen that the activity of both groups was falling off about equally previous to the beginning of the corpus luteum injection, which was begun on March 4th.

TABLE 2

EXPERIMENTAL GROUP

Rat No.	40 days before the treatment with corpus luteum extract was begun				30 days of corpus luteum treatment		
1077	14400	16150	8950	11450	7000	5500	2150
1091	8100	6000	6150	6300	4250	4800	2500
1093	7150	5800	5700	4500	2400	3150	2000
1095	6600	4650	5850	3700	2250	3600	2300
1096	15950	12900	14800	9200	5750	4350	4300
1100	11850	7450	6000	8000	6550	7000	7500
1110	6600	4500	3500	2400	4300	3350	4700
1125	14250	12500	9250	7400	8450	6650	5050
1128	8150	5650	3950	4050	3900	4500	4400
1164	14450	9000	4000	6250	4300	7150	4100
1166	7100	8400	5200	6800	5200	5050	3000
1167	9100	9650	6900	5050	5700	6250	5050
1181	19350	14000	7750	9000	7200	9700	9750
3268	10250	11950	12600	9700	10250	8350	6250
3269	4000	9450	10400	8000	7700	6950	5800
Average	10500	9200	7400	6850	5700	5700	4550

CONTROL GROUP

1079	9900	7400	2700	2000	7000	7750	6000
1092	4550	4450	5900	2900	3950	4200	1400
1101	3250	3800	1700	2600	3300	3100	1650
1115	10000	8350	8050	6150	8150	6650	6500
1173	1700	950	750	1000	1500	1400	1000
1180	2050	1800	1100	1300	1000	1200	1250
1182	16600	16700	9600	4250	4450	1750	3500
1183	14150	15700	13650	10150	8400	7900	3550
3270	6400	6200	8700	10200	5800	8750	8950
Average	7600	7200	5800	4500	4800	4700	3700

Table 2, showing the average daily activity of each animal reckoned by ten-day periods. Also the average for each group.

At this point the controls show an increase, a thing to be expected at this time of year, according to our experience. For the first ten days after the beginning of the treatment the activity of the experimental group fell off, but thereafter it remained on a par with the controls. The fact that the cyclic character of the activity was unaffected by the corpus luteum leads to the belief that the diminished total activity for the first few days was due to some other cause, possibly soreness produced by introducing the hypodermic needle repeatedly into the skin of the inguinal region. Some other unknown factor was doubtless in

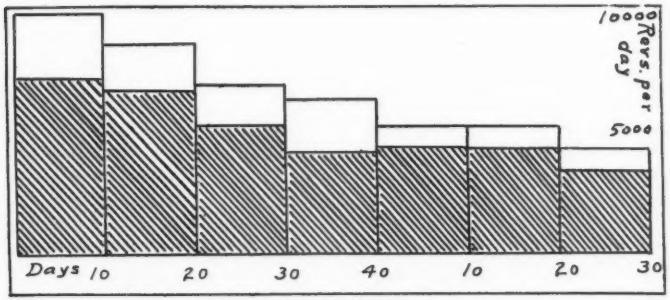

Fig. 2. The entire vertical rectangles, shaded and unshaded portions together, represent the total average activity of all of the treated animals for each of the respective ten-day periods. The shaded portions represent the activity of the controls.

part responsible for this condition, an examination of the individual activity records, not given in this paper for want of space, revealing that four of the fifteen subjects fell off extraordinarily in activity three or four days before the injections began and had not yet recovered their usual activity at that time. This fact, incidentally, does not vitiate the experiment because the periodicity of the activity was the main point at issue.

It is evident, then, that within the period of treatment and with the amount of extract used, and this was seemingly comparable to the amounts sufficient to produce sterility in experiments reported by other workers, the voluntary activity is unaffected. As bearing on this question, Knaus (2) has reported sterility which was only temporary, and that during continuance of the treatment pregnancies ultimately occurred which

resulted in unusually large litters. From this he infers that not development of ova but ovulation was for a time suppressed. Accepting the view of Allen and Doisy (8) that the female hormone is produced by the follicle cells under the influence of maturing ova, it is apparent that if the cyclic increase of activity is among the phenomena dependent upon the periodically secreted follicular hormone then under the conditions of this present experiment the luteal extract has not been antagonistic, at least in amount, to the follicular secretion proper, that is, the sex-hormone so-called. This view would not be inconsistent with that of Knaus referred to above. Papanicolaou (3) in a report published since the completion of this experiment states that (in the guinea pig) the full growth of the follicles and the process of ovulation are suppressed by the specific luteal hormone. Our results are not inconsistent with this view, especially the latter feature. They indicate, however, that although ovulation may not have occurred as usual, ova have been developing at the usual times, as shown by the continuance, throughout the period of treatment, of cyclic activity variations, and vaginal changes corresponding to the oestrual cycle. In a word, corpus luteum extract antagonizes ovulation, but does not prevent the development of ova.

SUMMARY

In an experiment with nine control white rats and fifteen others treated by subcutaneous injections of aqueous extract of corpus luteum over a period of thirty-seven days, the voluntary activity of the animals was unaffected in periodicity and total amount. Corpus luteum, therefore (assuming a potency of the extract used), does not prevent the formation of follicular hormone.

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THE EFFECT OF FOLLICULAR AND PLACENTAL HORMONES UPON THE MAMMARY GLANDS AND GENITAL TRACT OF THE OPOSSUM.

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It has been abundantly demonstrated in the last few years that the ovarian follicular hormone extracted by lipoid solvents from the liquor folliculi or similar extracts from the placenta* are capable of stimulating the genital tracts of immature or spayed adult female mammals.† The effect upon the mammary glands of such experimental animals has also been stated to be positive, but most authors refer to the mammary glands incidently and more or less casually, devoting chief attention to the size and vascularity of the uterus (Ascher and Grigoriu, 1911; Fellner, 1913, 1923; Iscovesco, 1914; Hermann, 1915). Vintemberger (1924) reports a well-executed set of experiments in which he induced an oestrous growth in the mammary glands of immature female and adult male rabbits by

*The name of Halban (1905) is especially associated with the theory of the placental hormone as the chief stimulus for the proliferation of the mammary gland during pregnancy. The theory, as stated by him, does not, however, deny the action of the ovary in the early part of pregnancy; the action of ovarian hormones in the early part of pregnancy is conceded.

†In a recent contribution Loewe (1925) has pointed out the superiority, in point of accuracy and definiteness, of the method of Allen and Doisy (1923), in which castrated mice or rats are used as test animals and the cornification of the vagina constitutes a positive result. Loewe generously states the method as being more accurate than his own, in which he used the swelling of the mammary glands (Hypermastie) as the diagnostic sign. See also Courrier (1925), Zondek and Aschelm (1925, 1926), Frank and Gustavson (1925).

means of intraperitoneal injections of crude bovine follicular fluid.§

For an experiment of this kind the opossum seemed to us to be well adapted, by virtue of the concentration of mammary glands in the limited area of the pouch and the fact that consequently, as Hartman (1923) has shown, changes in thickness in the gland are readily detectable, after some practice, by palpation. The same writer also showed, on anatomical grounds, that the mammary glands begin their growth under the influence of the growing graafian follicles.* It therefore seemed reasonable to suppose that, since in the opossum the mammary glands must needs be ready for lactation at a very early date (13 days or less after fertilization!), they might be unusually responsive to this hormone. At the same time the low position of the opossum in the mammalian series seemed to offer a further opportunity to test the species non-specificity of this hormone secured from man and other Eutheria (Cf. Allen, et al., 1924).

The experimental animals were all mature female opossums one, two, or more years old. All were castrated and the mammary glands allowed to involute until the glandular substance was no longer palpable, that is, until the skin containing the involuted glands had become as thin as skin elsewhere on the body. This condition is designated as "zero" in the protocols.† Growth in thickness or changes in turgescence of the glands were noted by palpation, and at frequent intervals vaginal smears were made by the Stockard and Papanicolaou

§The crude follicular fluid was employed by Sonnenberg (1907), Frank (1922), and others, and the crude "Press-saft" of organs was originally used almost exclusively. But it is desirable to avoid injection of toxic substances contained in so complex a mixture by precipitation of the toxic substances of the juice by means of (lipoid) solvents alcohol, acetone, and ether. This is accomplished in the method of Allen and Doisy, which follows in part the suggestive work of Fellner, Isovesco, and Hermann. Recently, Ralls, Jordan, and Doisy (1926) have worked out a simplification of the technique.

*The same conclusion had been reached in this manner by Robinson (1918) in his study of the oestrous phenomena of the ferret. Loeb and Hesselberg reported an initial but transient hypertrophy of the mammary gland in the guinea-pig, which they attributed to the Graafian follicles. Bucura (1909), through a priori reasoning, attributed the development of the secondary sex characters, as well as oestrus, to the follicle, perhaps the liquor folliculi itself, a conclusion which he suggested should be determined by experiment. The answer has now been definitely given by Allen and Doisy (1923).

†The ideal conditions for the experiment would have been to secure animals in anoestrum or the resting condition of December and early January, when the reproductive organs are considerably involuted to begin with. But it was impracticable to begin collecting the animals until the end of January, when most of them were pregnant or had young pouch young. Time then had to be allowed for complete involution to take place, palpation being the guide, according to the method referred to above (Hartman, 1919).

method, for in the opossum as in rodents, the histological condition of the vagina is a certain index to the changes in the uterus as well as the mammary glands, all standing in correlation to one another and to the ovary (Hartman, 1919). After autopsy the organs were fixed, imbedded, and for the most part sectioned and studied microscopically to determine the extent of regeneration effected or to observe the process in progress, as shown by mitotic cell division.

A word must be said about the vaginal smear in the majority of cases. In only two (Nos. 977 and 1000) was a typical clear-cut oestrous cornification of the vaginal mucosa obtained, with the almost complete disappearance of leucocytes and epithelial cells. In all other cases the latter never disappeared, although in all subjects cornification proceeded to a varying extent, perhaps more in one part of the organ than another (Hill and Fraser, 1925). Just such admixture of cells, giving an "atypical" picture to the vaginal smear, are quite common in animals experiencing abortive attempts at oestrous because of atresia of the graafian follicles (Hartman, 1924). Frank and Gustavson (1925, p. 6) doubtless had the same picture before them when they wrote: "Intermediate and partial reactions may occur." Such partial cornification will be referred to below as "atypical."

Two series of experiments were made. In six cases placental hormone, in five cases follicular hormone, was used. The hormone in all cases was prepared by Dr. Allen according to the method of Allen and Doisy and standardized in terms of rat units (Allen et al., 1924; Loewe, 1925). The injections were made twice daily, usually beginning with 0.5 cc. of the oil and increasing to 0.75 cc., 1.0 cc. and 1.5 cc. twice daily. In the notes the total amount injected over a period of days is recorded.

The essential details of the experiments are briefly stated in the following protocols.

PROTOCOLS

SERIES I. INJECTION OF PLACENTAL EXTRACT

No. 979, 1320 g. Feb. 11, both ovaries and uteri removed. Pouch young had been lost some time preceding operation.

March 3, mammary gland very thin, practically zero.

March 3 to 10 (20 days after operation), 9.0 cc. of placental hormone I injected (36 rat units).

March 9, mammary gland "thicker"; March 11, "decidedly thicker." Vaginal smear atypical, near-oestrous in character.

March 12, animal died. Organs considerably increased above castrate condition, but a great deal less than at normal oestrus.

No. 984, 885 g. (small). Ovaries removed Feb. 17. Pouch young had been lost some time before operation.

March 3-5, mammary glands almost zero.

March 5-9 (16 days after operation), 6.5 cc. placental hormone II injected (26 rat units).

March 9, mammary gland "rather thick"; March 10, "distinctly thick."

Mammary gland uniformly thickened. In section numerous thick-walled, regenerated alveoli with an occasional mitosis can be seen budding off of the more dilated, thin-walled alveoli of the previous lactation.

Uterus, vagina, and lateral vaginal canals enlarged and vascular, as in pro-oestrum. Uterus has well-preserved mucosa, including thickened epithelium and regenerated, rather closely packed glands, showing an occasional mitosis.

The growth changes in this experiment were carried well towards the oestrous condition.

No. 977. Pouch young removed February 19 and double ovariectomy performed.

March 10, mammary glands thin.

March 16-25 (25 days after operation), 16.75 cc. of placental hormone III injected (67 rat units).

March 21, mammary gland "thicker"; March 22, "clearly thicker"; cornified cells in vaginal smear.

March 23, killed.

Mammary glands very thick, as in pseudo-pregnancy of several days' standing (thicker than at normal oestrus). In section glands are seen to be well lobulated, each lobule consisting of numerous moderately dilated acini; mitoses numerous.

Uterus at least as at oestrus; vascular, round, and turgid; glands greatly convoluted; mitoses moderately numerous.

Vagina greatly hypertrophied; mucosa at end of post-oestrous desquamation; much cornified material in lumen; beginning of leucocytosis.

Lateral vaginal canals collapsed, as normally in metoestrum, giving evidence of previous great dilatation.

Altogether, the reproductive organs present the picture of an animal well advanced in the pseudo-pregnant condition.

No. 981. Embryos near term February 16; both ovaries and both uteri removed. February 24, mammary gland involuted, thus: March 3, "just palpable." March 9, 16, 19, "very thin."

March 27-31 (39 days after operation), injected 6.0 cc. (24 rat units) or the remaining portion of placental hormone III (partly used on No. 977).

April 1, mammary gland decidedly thicker. Vaginal smear "atypical" oestrous type.

April 3, glands thick and firm, as in late pro-oestrum.

April 7, killed. Organs again greatly though not completely involuted, autopsy having been made 7 days after discontinuing the injections.

No. 993(a), 2000 g. Pouch young removed and female ovariectomized February 27.

March 21 (22 days after operation), mammary glands zero.

March 21-28 (interval 24 days), 15.25 cc. placental hormone IV injected. No effect whatever was noted, either in the vaginal smear or on the mammary glands. The results being clearly negative, the animal was kept for another experiment (v.i.),

The hormone here used was extracted from a human placenta which had inadvertently stood in the warm sun for a whole day. Putrefactive agents or autolysis or both had doubtless rendered the hormone inert.

No. 1000. Trapped at Columbia, Mo., and ovariectomized on February 4.

February 25 to March 5, 17.0 cc. of placental hormone V were injected (69 rat units).

Mammary gland very thick, as in early pseudo-pregnancy. Considerable development of new alveoli, moderately dilated, mostly one-layered; mitoses numerous.

Vagina greatly enlarged, almost maximal, as at oestrous; desquamation of cornified layer almost completed. Moderate leucocytosis of vaginal epithelium.

Lateral vaginal canals give every appearance of collapse after great oestrous dilatation.

In this experiment the hormone action clearly resulted in growth and hypertrophy of the organs to a degree normally found in early pseudo-pregnancy.

SERIES II. INJECTION OF FOLLICULAR EXTRACT

No. 957, 1118 g. Ovariectomized January 21 while in anoestrum; one ovary transplanted in pieces into a rectus abdominis muscle. January 27, no change in mammary gland. Escaped end of January; accidentally recaptured March 23. April 3, mammary glands reported in cage notes as temporarily somewhat thickened; vaginal smear, however, showed no cornified cells. No further sign of sexual activity whatever.

May 11 to 16 (110 days after operation), 6.25 cc. of follicular hormone injected (18.75 rat units).

May 17, atypical pro-oestrous smear. Killed May 19. Mammary gland somewhat thickened.

Uterus as in early pseudo-pregnancy; mitoses very numerous in greatly convoluted glands, which are well ciliated.

Vagina in later stage of cornification with great infiltration of leucocytes, as in metoestrum.

This experiment may have been complicated by the possible influence of the transplant, which, however, could not be found at autopsy.

No. 982. Pouch young 5 or 6 days old; removed, and female ovariectomized February 17.

March 27. Mammary glands zero.

May 11 to 16 (93 days after operation), 9.25 cc. follicular hormone injected (18.5 rat units).

May 17. Vaginal smear oestrous, atypical. No proliferation of mammary gland, only congestion and some extravasation. No other record made.

No. 958. Oestrus January 27; February 11. dioestrum.

February 22. Ovariectomy; mammary gland nearly zero.

May 21 to 26 (88 days after operation), 14.5 cc. follicular hormone injected (43.5 rat units).

May 29, killed. Mammary gland slightly thickened, firm. Uterus as in early pro-oestrum. Lateral vaginal canals slightly, median vagina considerably enlarged; vaginal smear pro-oestrus, atypical.

This experiment resulted in a correction of the castration atrophy of three months' standing, but failed to advance the organs beyond the early pro-oestrous condition.

No. 987, 1116 g. Six pouch young, 10 days old, removed and female ovariectomized February 20.

May 21 to 31 (90 days after operation), 32.0 cc. follicular hormone injected (96 rat units).

June 1, killed. Vaginal smear more or less as at oestrus, but atypical. Vagina and lateral vaginal canals about as in dioestrum; uteri and mammary glands developed to the normal oestrous stage.

A large quantity of hormone was used in this experiment. The subject recovered thoroughly from its profound castration atrophy. Uteri and mammary glands had developed well towards the oestrous condition, but the vagina and the lateral vaginal canals, normally the first to respond, remained the most refractory.

No. 993(b), 2000 g. (very large). This animal had received placental hormone IV (v.s.) without effect March 21 to 28.

May 23 to 29 (85 days after operation), 18.0 cc. (90 rat units) follicular hormone injected. Killed 4 days later.

Mammary glands slightly thickened; regeneration of alveoli, numerous mitoses. Genital tract recovered from castration atrophy of preceding months, but only slightly advanced towards oestrous condition.

DISCUSSION OF RESULTS

It is seen from these experiments that in all cases but one (No. 993, first injection), the hormone stimulated the growth of the mammary glands and the genital tract and corrected the atrophy to which these organs are always subjected after ovariectomy. In certain cases, especially in Series II, this is as much as can be said for the action of the hormone; but in others, notably in No. 997 and No. 1000, of Series I, not only were the phenomena of oestrous pronounced and quite typical, but the organs went farther and exhibited distinct symptoms of pseudo-pregnancy. This holds especially for the mammary

TABLE 1

Giving Summary of Data Presented in the Protocols

SERIES I: PLACENTAL EXTRACT

No. of animal	Date of		Time interval (days)	Amount injected (cc.)	Rat units	Results
	Castration	Injection				
979	Feb. 11	Mar. 3–10	20	9.0	36	Fair
984	Feb. 17	Mar. 5–9	16	6.5	26	Very good
977	Feb. 19	Mar. 16–25	25	16.75	67	"Pseudopregnant"
981	Feb. 16	Mar. 27–31	39	6.0	24	Fair
993	Feb. 27	Mar. 21–28	22	15.25	0	Negative
1000	Feb. 4	Feb. 25–Mar. 5	21	17.0	69	"Pseudopregnant"
SERIES II: FOLLICULAR EXTRACT						
957	Jan. 21	May 11–16	110	6.25	18.75	Good
982	Feb. 27	May 11–16	73	9.25	18.5	Poor
958	Feb. 22	May 21–26	88	14.5	43.5	Fair
987	Feb. 20	May 21–31	90	32.0	96.0	Fair
993	Feb. 27	May 23	85	18.0	90.0	Good

gland, but is also true to a large degree of the genital tract as well.

The hormones from both sources (human placenta, pig's graafian follicles) thus had the expected effect. However, it was somewhat surprising, in view of former work on the rat, that the extract of human placenta should turn out to be so much more potent than the extract of the pig follicular fluid.

The explanation of this discrepancy, if such it is, lies, it seems to us, in the extent of castration atrophy attained by the organs in the two series, much more profound in the second series, which proved to be the more refractory. In this series 110, 73, 88, 90, and 85 days, respectively, elapsed between the removal of the ovaries and beginning of the injections, against 20, 16, 25, 39, 22, and 21 days for the first series (Table 1). It is a well-known fact, holding true for the opossum as well as for the classical experimental mammals and man, that time is the determining factor in the extent of castration atrophy. Gynecologists are furthermore well aware of the refractory nature of the hyperinvoluted puerperal uterus which sometimes accompanies lactation. This condition and other forms of amenorrhea are doubtless due to ovarian insufficiency, as has been suggested by various gynecologists (e. g., Novak and Graff, 1921; Guggisberg, 1925). The symptoms in such cases simulate those of complete castration. Any one who has had any considerable experience with experimental mammals knows the almost invariable correlation between atrophy of the genital tract and mammary gland and hypotypical ovaries.

Guggisberg (1925) offers a somewhat different explanation to the refractiveness of the uterus after a lapse of time. He contends that the sensitization of the uterus by the ovary is prerequisite (he reports almost no effect on the mammary glands in any case). This effect wears off more and more after castration, until the uterus becomes permanently refractory. As a matter of fact, Guggisberg states that, for obscure reasons, a considerable percentage of animals fail to react, whether castrated or not.

The two series of experiments presented are, therefore, not strictly comparable. Whether the factor of time, as set forth above, is the true explanation of the differences in results attained must be left for further experimentation. The matter is of considerable importance if the method is to be used in the standardization of the hormone.

As a further result of the foregoing experiments, the species non-specificity of the sex hormone is thus extended to the marsupial: extracts of human placenta and pig's liquor folliculi are found effective on the reproductive organs of a representa-

tive of the lowest class of mammal having an intrauterine gestation. This result is, of course, not surprising, if one consider that the ovaries of birds (Allen, et al., 1924) and fishes (Fellner, 1925) furnish the hormone in question. It would seem probable, therefore, that this ovarian hormone consists of a definite compound, or at least a common organic radicle, widely distributed in the animal series. In this sense this hypothetical body is comparable to such fundamental chemical entities as adrenalin, insulin, and iodothyrin (thyroxin), which are widely distributed.

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EPHEDRIN THERAPY IN ADDISON'S DISEASE *

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Our knowledge of Ma Huang, according to Chen, dates back into the dim history of China some 5,000 years. "Ma" signifies astringent, and "Huang" yellow. The first mention of it appeared in the Chinese dispensatory in 1596 A.D. Nagai, in 1887, isolated an alkaloid from it which he named ephedrin. The recent work of Chen and Schmidt has focused attention on the pharmacologic actions of ephedrin and its possible clinical value.

The chemical structure of ephedrin, according to Chen, is probably C_6H_5OH-CH. CH_3-NHCH$_2$. Kendall, of the Mayo Clinic, has investigated the fundamental behavior of ephedrin and epinephrin, particularly in relation to their oxidizing properties. In this connection we quote him as follows:

"The structural formula of epinephin has been established by its synthesis. The chemical groups present and their precise arrangement are known beyond question. The chemical structure of ephedrin is also known, and inspection of the structural formulas of these two substances show them to be closely related. The activity of each is bound up in the actively functioning groups which probably are the methylamin group in ephedrin. The difference in the physiologic activity of these two substances lies in the presence of the two hydroxyl groups in the benzene ring of epinephrin. These two groups which are adjacent carry with them the assurance of the short time for activity of the molecule, because through these groups the molecule is easily destroyed. The benzene ring in ephedrin is unsubstituted, and in vivo and in vitro experiments show that the molecule is very much more stable than is epinephrin. Epineph-

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rin is oxidized comparatively easily, owing to the presence of the two hydroxyl groups of the benzene ring. Ephedrin is stable towards oxidizing agents. Physiologic experiments have shown that epinephrin functions almost instantly and its action is over within a few minutes, but the effect of ephedrin lasts for several hours. The physiologic dose of epinephrin is measured in tenths of milligrams and the physiologic dose of ephedrin is many times as great."

The pharmacologic action of ephedrin has been carefully investigated by Amatsu, Kubato, Chen and Schmidt. In experimental animals, dogs, cats and rabbits, ephedrin has an effect on smooth muscle, on secretion, and on the circulation, indicating sympathetic stimulation similar to that of epinephrin. There is a prolonged rise of blood pressure with cardiac acceleration and frequently vasoconstriction. The heart effects are evinced by increased rate and strength of contraction which is not inhibited by atropin or by sectioning the vagi. Repeated injections in the animal cause a lowering of blood pressure. Chen's investigation indicates that ephedrin produces a stimulation of the accelerator ganglia of the heart and the accelerator nerve endings and depression of the heart muscle. Its vasomotor action is due to stimulation of the vasomotor nerve endings.

The toxicity of the drug is low; 25 mg. given subcutaneously to rabbits in daily doses for four weeks is without toxic effects. The total dosage here consumed was eight times the single minimal lethal dose. No tolerance developed to the mydriatic or pressor action of the drug. No visceral damage was discovered.

Because of the epinephrin-like action and the low toxicity, Chen suggested its use in states of low blood pressure, shock and hemorrhage, in Addison's disease and other types of hypotension. Because of its localized vasoconstrictor action on the nasal mucous membrane, attention was called to its possible usefulness in this field. The outstanding advantages of ephedrin, as compared to epinephrin, are its more prolonged action and its effectiveness when given orally. Miller has reported the results obtained by the use of ephedrin in a group of eighty cases representing various pathologic conditions. No significant toxic effects were observed when doses of from 50 to 125 mg. were given by mouth or subcutaneously. In a large majority of the cases there was some elevation of blood pressure. In 20 per

cent the increase was 40 mm. of mercury or more. The increased pressure was maintained for from three to eight hours. In two cases of Addison's disease there was some symptomatic improvement. Relief was obtained in the attacks of bronchial asthma, and temporary improvement was noted in a case of circulatory collapse. An increase of the basal metabolic rate was observed in two cases. Miller states that the minimal lethal dose of ephedrin is from thirty to one hundred times that required to produce physiologic effects.

In the Mayo Clinic the drug has been used in several different connections. Gaarde and Maytum report that in twenty-six cases of active ragweed fever, treated daily with ephedrin (65 mg. capsules), one-half of the patients obtained almost complete relief for from three to seven hours following each administration. In seven cases there was no effect, and four patients had but moderate relief. On the other hand, their results in a limited study of bronchial asthma were not so satisfactory. In treating the nose and throat, Hempstead has used ephedrin for constriction of the nasopharyngeal mucous membrane and finds that its effect persists much longer than that of epinephrin, and is not followed by the compensatory dilatation so often seen when epinephrin is used. He has also used it in combination with local anesthetics in tonsillectomy. While he has not yet worked out to his entire satisfaction the exact equivalent dosage, he believes that ephedrin promises relief from the dilatation and subsequent bleeding, which has been a considerable menace when epinephrin has been employed.

Our studies cover the use of the drug in a series of twenty-eight cases: fourteen of Addison's disease, two of questionable Addison's disease, a control group of nine cases of hypotension, two cases of pituitary tumor, and one case of endocrine obesity. Fourteen patients presenting the classical picture of Addison's disease were treated in the hospital. Three were given a daily dose of from 50 to 250 mg. of ephedrin for prolonged periods of time, either with or without the Muirhead regimen. The blood pressure was taken twice daily. In three cases repeated tests with ephedrin were made in which a single dose of 50 or 70 mg. was given orally under basal resting conditions; the blood pressure and pulse rate were taken at half-hour intervals

for a period of three or four hours. Eight patients were given one or more doses of 50 or 70 mg. of ephedrin, but the blood pressure and pulse rate were not followed systematically. Three patients, during the administration of the Muirhead treatment, were observed in the shock-like condition which occurs at times in the decompensated stages of Addison's disease; they were given ephedrin in the hope of tiding over a crisis. In addition, two questionable cases of Addison's disease, with mild generalized pigmentation, low blood pressure and weakness, were given ascending doses of ephedrin, up to a maximal dose of 300 mg., without toxic effects.

The calorigenic action of ephedrin was studied in three cases of Addison's disease.* From 50 to 100 mg. of ephedrin was given orally, and the basal metabolic rate was determined at half-hour intervals for a total of four or five determinations.

Nine cases of chronic nervous exhaustion and hypotension were used as controls, and the clinical, subjective and objective effects of ephedrin studied. Two cases of pituitary tumor with low blood pressure and low metabolic rate were treated with ephedrin and thyroid extract as a preoperative measure.

EFFECTS ON BLOOD PRESSURE

Addison's Disease. The effects of repeated increasing doses of from 25 to 100 mg. of ephedrin are shown in three cases in Table 1. In Case 1 comparative determinations were carried out with small and large doses of ephedrin and also (for a control) without the drug. With 25 mg. a demonstrable rise of 20 mm. of mercury was noted five hours later, and the elevation was maintained to some degree for a period of four hours. There was no change in the diastolic pressure. A similar test on the following day produced no changes in the blood pressure and a subsequent test with 250 mg. showed no significant effect on the blood pressure. Case 2 showed a moderate response after 60 mg. of ephedrin on two successive days. A maximal increase of 18 mm. of mercury of at least two hours' duration was noted, without significant change in the diastolic pressure. In Case 3 there was an increase in the systolic pressure of 20 mm. of mercury in one hour and fifteen minutes after the administration of from 50 to 60 mg. of the drug. The diastolic

*This was attempted in other cases but the strain resulting from repeated breathing tests necessitated its discontinuance on several occasions.

TABLE 1

Effects of ephedrin on blood pressure in cases of Addison's disease

Case	Date, 1925	Time	Ephedrin, mg.	Blood pressure	
				Systolic	Diastolic
1	4–29	9:00	25	92	58
		10:00		100	60
		11:00		100	60
		12:00		90	56
		1:00		100	60
		2:00		112	70
		3:00		100	60
		4:00		104	56
		5:00		98	62
		7:00		98	60
		8:00		98	56
	4–30	9:00	25	92	58
		10:00		90	60
		11:00		96	54
		12:00		82	50
		1:00		86	50
		2:00		86	50
		3:00		90	58
		4:00		92	60
		5:00		92	60
		6:00		90	60
	5–2	9:00	None	86	56
		10:00		80	58
		11:00		122	70
		12:00		88	32
		1:00		88	58
		2:00		88	56
		3:00		88	50
		4:00		90	52
		5:00		95	60
	5–4	11:00	250	88	54
		3:00		94	
		5:00		98	64
2	1–8	15 minutes	30	Patient at rest	
		10:50		110	68
		11:35		100	70
		12:05		108	68
		1:05		106	68
		1:35		106	68
		2:05		108	66
	1–9	9:00	60	108	74
		9:00–9:15		Patient at rest	
		10:00		112	62
		11:00		118	72
		11:30		118	70
		12:00		114	66
	1–11	9:00	60	100	60
		9:15			
		9:30		100	60
		10:00		112	68
		10:30		112	68
		11:00		112	65
		11:30		118	70
		12:00		118	70

TABLE 1 (Continued)

Case	Date, 1924	Time	Ephedrin, mg.	Blood pressure	
				Systolic	Diastolic
3	12–20	4:45	50		
		5:00		115	70
		5:35		124	58
		6:00		140	74
		6:30		140	70
		7:00		138	58
		7:30		136	52
		8:00		120	48
		8:30		110	46
		9:30		84	40
		10:00		86	40
	12–21	7:20	50	100	68
		7:35		104	64
		8:05		104	58
		8:35		120	68
		8:50		116	58
		9:00		114	56
		10:00		118	60
		10:30		114	54
		12:00		106	54
		2:00		108	58
		6:00		108	58
	1925 1–2	10:15	60	124	58
		10:30			
		10:45		120	56
		11:15		142	60
		11:45		144	62
	1–15	10:15	60	124	58
		10:30			
		10:45		120	56
		11:15		142	60
		11:45		114	62

pressure showed no significant change. Subsequent comparative tests carried out on successive days showed comparable increases in blood pressure.

The effects of the prolonged administration of daily doses of from 50 to 250 mg. of ephedrin on the blood pressure in Addison's disease is shown in Figures 1, 2 and 3. The administration of ephedrin is associated with a slight rise in blood pressure (Fig. 1). The fall of pressure on the seventh day was thought to be due either to overexertion or to catharsis. Bramwell has emphasized the ill effects of catharsis. This patient confirms his observation since he himself ascribes his relapse to purgation. Figure 2 shows a sustained increase in blood pressure over a period of five days, under the influence of ephedrin. After the administration of ephedrin was stopped, the diastolic pressure decreased but the systolic pressure was sustained. The comparative effects of ephedrin and another member of the

epinephrin group are shown in this figure. Figure 3 is of unusual interest in that it depicts the increase in both systolic and diastolic pressure following the use of 250 mg. of ephedrin daily for fourteen days. Despite this increase in blood pres-

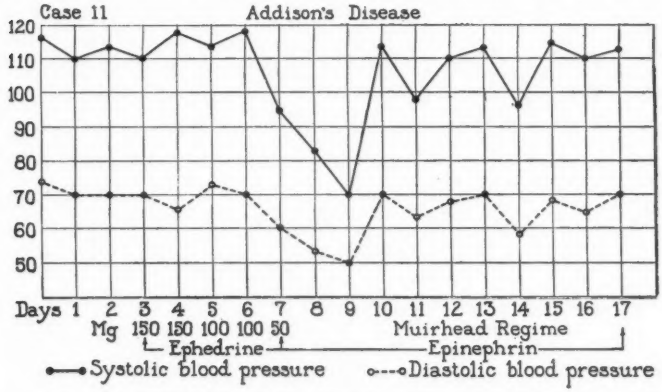

Fig. 1. A marked fall in blood pressure on the seventh day, attributed to catharsis or overexertion. The pressure had been satisfactorily sustained by ephedrin up to that time.

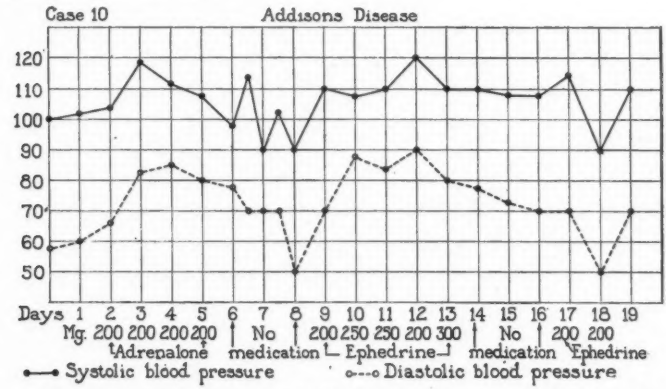

Fig. 2. A sustained increase in blood pressure over a period of five days, after the administration of ephedrin.

sure, the patient died under treatment. The final drop in blood pressure is regarded as an antemortem phenomenon.

A third group comprises eight cases of Addison's disease in which one or more doses of ephedrin were administered for

its clinical effect, but without such detailed studies of the effect on blood pressure or metabolism. Some patients in this group were under ephedrin treatment over a number of days. In three of these cases, ephedrin was employed more or less as a last resort in the effort to tide the patient through a critical period of the disease. The clinical results in advanced cases of Addison's disease were distinctly disappointing. In not a single such instance did we feel that ephedrin contributed much to the feeling of well-being or to the recovery of the patient.

In two cases of questionable Addison's disease increasing doses of ephedrin were administered up to 300 mg. a day. The effects on blood pressure were transitory and the general level

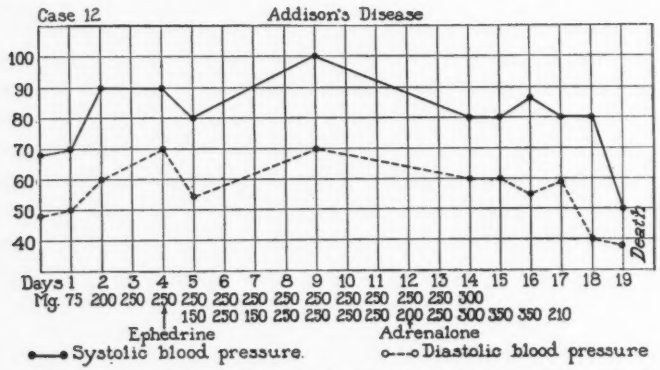

Fig. 3. An increase in both systolic and diastolic blood pressures after the daily use of ephedrin for a period of fourteen days.

of blood pressure was not significantly increased during a period of five days.

Nervous Exhaustion and Hypotension. The effects of ephedrin on the blood pressure in nine cases of hypotension are illustrated by typical cases in Tables 2 and 3. There were moderate increases in the systolic pressure ranging from 10 to 30 mm. of mercury. The increase in diastolic pressure was not consistent, but averaged about 11 mm. of mercury. The maximal changes in blood pressure occurred from one-half to three hours after the administration of the drug and persisted from three to six hours. Objectively, there was no other evidence of clinical improvement, although several of the patients were convinced that they felt somewhat stronger.

TABLE 2

Effects of ephedrin on blood pressure in cases of chronic nervous exhaustion

Case	Date, 1925	Time	Ephedrin, mg.	Blood pressure		Pulse	Remarks
				Systolic	Diastolic		
1	2-5	2:45		102	62		
		3:00	75				
		3:30		100	56		
		4:00		110	58		Weakness and trembling
		4:30		112	72		
		5:00		118	56		
		5:30		118	58		
		6:00		110	60		
	2-6	4:00		102	54	92	
		4:15	75				
		5:15		118	70	72	
		5:45		126	70	74	
		6:15		122	70		
		6:45		104	48	92	Weakness and trembling
		7:15		112	64		lasting four hours
2	2-7	12:45		95	55	80	
		1:00	75				
		1:30		94	58	88	
		2:00		94	62	88	
		2:30		98	60	100	
		3:10		80	45		
		3:30		94	60		
		4:00		120	60	84	
3	4-20	10:25		100	70		
		10:40	75				
		11:10		110	70		
		11:40		110	78		
		12:40		104	58		
		1:40		98	50		

TABLE 3

Comparative effects of ephedrin and epinephrin in a case of chronic nervous exhaustion

Date 1925	Time	Drug administered and amount	Blood pressure		Pulse	Remarks
			Systolic	Diastolic		
2-24	2:30		120	60		
	2:45					
	3:15		118	70		
	3:45	Ephedrin, 70 mg. orally	115	50	88	
	4:15		150	75	80	Dizziness
	4:45		150	70	72	
	5:15		142	70	76	Nausea
	5:45		138	70	72	
2-26	9:20		105	70	92	
	9:35		118	60	96	
	9:50	Epinephrin, 0.5 cc. hypodermically	118	60	88	
	10:05		110	56	96	
	10:20		106	50	96	
	10:35		105	55	96	

Pituitary Disease. In the first case of pituitary disease, in response to single doses of 50 mg., the effects on the blood pres-

sure were not decisive. In Case 1, Table 4, 100 mg. of ephedrin was given daily for a total dosage of 3.05 gm. (Fig. 4). There was a gradual and steady increase in the systolic and diastolic pressures. Thyroid extract was given simultaneously and the increase in blood pressure was accompanied by an increasing

TABLE 4

Effects of ephedrin on blood pressure and basal metabolic rate

Case	Date, 1926	Ephedrin, mg.	Time after giving of ephedrin, minutes	Basal metabolic rate	Blood pressure	
					Systolic	Diastolic
1 (Pituitary tumor)		50 (orally)		— 6	100	56
			15	—11	100	58
			30	— 8	102	56
			60	— 2	108	60
			90	— 1	106	54
2 (Chronic exhaustion syndrome after pregnancy)	3–30	50 (orally)		— 2		
			15	— 2		
			30	— 4		
			60	— 1		
			90	+ 5		
	4– 1	50 (orally)		—14		
			15	—10		
			30	— 8		
			60	— 7		
			90	— 7		
	4– 7	50	9:15		98	66
			9:45		104	74
			10:15		110	72
			10:45		100	60
			11:15		100	60
			11:45		122	64
			12:15		120	76
			12:45		100	60

metabolic rate (Fig. 4). The clinical improvement in this patient was not sufficient to justify surgical intervention.

EFFECTS ON PULSE RATE

No significant variations were observed in the pulse rate following the administration of ephedrin alone. About an equal number showed an increase or a decrease from 10 to 15 beats a minute. In one case of Addison's disease, following a rise of systolic pressure to 176 mm. of mercury, the pulse became bounding in character and slightly irregular; the rate was 46.

CALORIGENIC EFFECTS

The calorigenic effects of ephedrin were determined in three cases of Addison's disease, from 50 to 100 mg. of ephedrin being given orally, and the basal metabolic rate determined from

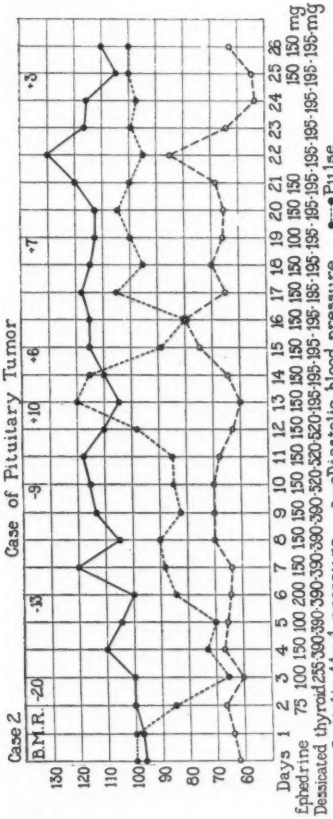

Fig. 4. A gradual increase in systolic and diastolic blood pressures after a total dosage of 3.05 gm. of ephedrin. With the simultaneous administration of thyroid extract an increase in basal metabolic rate is also noted.

four to six times at intervals of a half hour (Table 5). In Case 1 there was a drop of 12 points in the basal metabolic rate. In Case 2 there was an increase from —4 to +2. In Case 3, 100 mg. of ephedrin was given, and the basal metabolic rate increased from —27 to —17 after a period of one hour and twenty minutes.

In a case of questionable Addison's disease, the administration of 150 mg. of ephedrin was followed by an increase in the metabolic rate of from +2 to +12. This rise occurred in two hours (Case 4, Table 6).

TABLE 5

Calorigenic effects of ephedrin in cases of Addison's disease

Case	Ephedrin, mg.	Time after giving of ephedrin, minutes	Basal metabolic rate
1	50		+ 9
		30	— 3
		60	+ 3
		90	+ 4
		120	+ 5
2	50		— 4
		15	— 4
		30	— 6
		60	+ 2
		90	+ 2
3	100		—27
		80	—17

TABLE 6

Calorigenic effects of ephedrin in cases with chronic exhaustion syndromes

Case	Ephedrin, mg.	Time after giving of ephedrin, minutes	Basal metabolic rate
1 (Obesity)	50		+ 2
		15	— 8
		30	— 5
		60	— 7
		90	— 9
2 (Exhaustion syndrome)	50		+ 1
		15	+ 4
		30	+ 2
		60	+ 7
		90	— 1
3 (Chronic exhaustion)	50		—11
		15	— 6
		30	—13
		60	— 1
		90	+ 4
4 (Questionable Addison's disease)	150		+ 2
		60	+ 9
		120	+12
		180	+ 6

The calorigenic effect of ephedrin was likewise determined in four cases of hypotension, with dosages of 50 mg. orally. A slight increase was demonstrable in all cases after a period of one hour. The effects seemed quite transitory and one hour

afterwards there was a tendency for the metabolic rate to decrease. The patient with endocrine obesity (Case 1, Table 6) showed a decrease of 11 points with 50 mg., but with 100 mg. there was an increase of 13 points in three hours. In the cases of pituitary tumor there was a demonstrable increase of the basal metabolic rate after 50 mg. of ephedrin had been given.

TOXIC EFFECTS

The subjective symptoms from ephedrin in doses of 50 to 100 mg. were remarkably slight. Three patients with Addison's disease manifested reactions which may have been related to the administration of ephedrin. In one case 150 mg. of the drug was administered twice daily while the full Muirhead treatment was being given. On the fourth day ephedrin was taken soon after 0.5 cc. of epinephrin had been given subcutaneously. A throbbing headache and profuse perspiration resulted, and the systolic blood pressure increased to 176 mm. The diastolic pressure was 68. The pulse was irregular and the rate dropped to 46. The reaction subsided in five minutes. No similar reactions were noted following the administration of either drug alone. It was felt, therefore, that the combination of the two drugs was probably responsible for the unusual reaction. In the second case of Addison's disease there was a slight gastric upset which may have been due in part to overexertion. The third patient complained of weakness and trembling of a transitory nature after 100 mg. of the drug had been given. Some of the patients with questionable Addison's disease manifested weakness, marked trembling, and palpitation thirty minutes after taking 150 mg. of ephedrin. The effects lasted about thirty minutes and were not accompanied by significant changes in the blood pressure or pulse rate.

The patients with hypotension seemed less tolerant to ephedrin than those with Addison's disease. Three complained of palpitation, weakness and trembling, within thirty minutes after taking the drug. In one case this persisted for four hours. These reactions were mild, and the trembling and tremor were subjective rather than objective in character. The patient with

pituitary tumor tolerated the prolonged administration of the drug remarkably well and no toxic symptoms were observed. Mild trembling was noted by practically all ambulatory patients after doses of 65 mg. or more. Patients at rest appeared to tolerate this drug better than those who were ambulant.

REACTIONS OF THE SKIN CAPILLARIES

Ephedrin in 10 per cent concentration in physiologic solution of common salt was used in determining the skin reaction. A drop of the solution was placed on the skin of the forearm, and several bloodless punctures were made through it with a needle. Control tests with 1:1000 epinephrin solution and with trauma alone were made. The ephedrin reaction consists of a small red central area, about 3 to 6 mm. in diameter, surrounded by a patchy white border which is quite irregular and not constant. Reflex erythema frequently appears. The epinephrin reaction consists of a small white central area surrounded by one of erythema. The trauma reaction usually consists of a small central red area with reflex erythema but no white border. The ephedrin reactions were inconstant and the capillary contractions less decisive than with epinephrin. In one case of Addison's disease the zone of constriction, or white area, was absent with ephedrin, the red center alone appearing. Further work is being carried out in this connection.

GENERAL CLINICAL EFFECTS

In cases of Addison's disease the results from the use of ephedrin are distinctly disappointing. We have seen no evidence of significant improvement in patients with advanced Addison's disease. Certainly the resulting moderate increase in blood pressure was not accompanied by a marked feeling of well-being, significant increase in strength, or relief from the gastric symptoms or circulatory asthenia. The subjective improvement following a Muirhead treatment is much more pronounced than that from the use of ephedrin.

Full therapeutic doses of ephedrin resulted in some subjective improvement in certain instances. An occasional patient

admitted feeling somewhat stronger for several hours following its administration. This, however, was an exception. Full therapeutic doses usually resulted in minor toxic manifestation, palpitation, and a peculiar subjective sensation of weakness and trembling.

In one case of early Addison's disease excellent clinical results were obtained, comparable in every way to those we have seen in similar cases under the Muirhead regimen. The feeling of weakness and exhaustion disappeared and the patient felt buoyant, strong and refreshed. However, the treatment was without significant effects in other early cases of Addison's disease.

In hypotensive and hyposthenic states not due to Addison's disease, ephedrin may prove of value at times. A more extensive use is advocated in this connection.

CONCLUSIONS

1. Ephedrin is relatively nontoxic and may be used orally, subcutaneously, or intramuscularly without undue toxic effects, in amounts sufficient to increase blood pressure and to raise the rate of metabolism.

2. While increase in blood pressure and in rate of metabolism may be demonstrated in Addison's disease as the result of ephedrin, these changes are rarely accompanied by significant subjective or objective evidences of clinical improvement.

3. Ephedrin is not an effective specific remedy in the treatment of Addison's disease. The results obtained with ephedrin have been disappointing and do not compare in value to those obtained by the Muirhead treatment.

4. Because of its low toxicity and its effect on blood pressure and on metabolism when administered orally, its further trial in other hypotensive and hyposthenic states is warranted.

5. It must not be concluded from the results reported herein that ephedrin will not prove useful in other fields; the references suggest that it will.

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A CASE OF DIABETES INSIPIDUS ASSOCIATED WITH SYPHILIS OF THE HYPOPHYSIS *

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Diabetes insipidus is a clinical syndrome characterized by polydipsia and polyuria. A distinction is frequently made between primary or essential, and secondary or symptomatic polyuria. In the primary variety, polydipsia and polyuria are the chief clinical symptoms; in the secondary variety, these symptoms constitute only part of the clinical picture, which is usually that of some organic brain disease. The volume of urine in diabetes insipidus varies from a slight increase above the normal to enormous quantities in twenty-four hours. The polydipsia varies with the polyuria.

Numerous clinical studies have been made of diabetes insipidus. These have brought out a number of facts but have led to a great many more speculations. Some writers have rightly termed diabetes insipidus a disease of theories. The urine in diabetes insipidus has the following characteristics: The volume is markedly increased, it is pale in color, of low specific gravity, and poor in salts as indicated by freezing-point determination. The ordinary tests of renal function have usually shown no deviations from normal, in uncomplicated cases.

Complete agreement as to the pathologic physiology of diabetes insipidus has not been reached. The idea that the kidneys lose their concentrating capacity has been generally abandoned [Meyer and Meyer-Bisch (1)]. On the basis of metabolic studies, Veil (2) maintains that diabetes insipidus is a disturbance in intermediary metabolism of water and sodium chloride. He believes that the entire organism is adjusted to a lower niveau of osmotic pressure which is the starting point of the insipidus. Meyer and Meyer-Bisch conclude that a hypotonic sodium chloride solution not only circulates through the kid-

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neys in the blood, but that the tissues yield a hypotonic solution to the blood. There seems to be a disturbance in exchange between the tissues and blood as well as between the blood and kidneys.

I have been able to collect from the literature the records of 105 cases of diabetes insipidus with autopsy findings. The pathologic lesions held responsible divide themselves into three distinct types, depending upon the prevailing theories and experimental production of polyuria. The first period was influenced by Claude Bernard's classical experiments (3) in which he demonstrated that piqure of the floor of the fourth ventricle led to polyuria, and lasted until 1912 when Frank (4) directed attention to the frequent association of diabetes insipidus with signs of hypophysial lesion. The second period may be termed the hypophysial period and extends from 1912 to 1920. During this period special attention was directed toward the hypophysis, other lesions being accorded secondary importance, and when gross lesions were absent, it was maintained that pressure upon the hypophysis by distant lesions might cause polyuria. The third period is again based upon experimental physiology and finds its origin in the work of Camus and Roussy (5), who showed that injury to the floor of the third ventricle near the insertion of the infundibulum leads to polyuria. There is a distinct tendency at the present time to minimize the importance of lesions of the hypophysis and revert to the cerebral idea of the etiology of polyuria.

Leschke (6) was among the first to maintain that diabetes insipidus is due to a lesion at the base of the midbrain, and based his arguments upon a large amount of clinical and experimental material. He gives protocols of 16 cases in which there was complete destruction of the hypophysis without polyuria. A careful study of such cases was made by v. Hann (7), who compared them with cases of diabetes insipidus in which pathology of the hypophysis was found. He concludes that the occurrence of polyuria depends not only upon the condition of the posterior lobe, but on that of the anterior lobe as well. Disturbances of the posterior lobe with an intact anterior lobe lead to diabetes insipidus; if the anterior lobe is injured at the same time as the posterior lobe, diabetes insipidus does not develop. Extension of the process to the anterior lobe may cause

an existing diabetes insipidus to disappear. This would account for such cases as Simmonds (8) reports in which a metastasis from a carcinoma of the breast produced polyuria which later disappeared.

That lesions of the base of the midbrain do not invariably produce polyuria is abundantly proved by reports of cases, usually tumors of the hypophysial anlage, which involve this region not associated with polyuria.

REPORT OF CASE

The patient, an Austrian garment worker, was admitted to the Cook County Hospital, on the service of Dr. Jos. Miller, August 17, 1915, and discharged on August 24, 1915. He was then 40 years old. His complaints were: incontinence of urine, weakness, polyuria, impotence, constipation and a constant feeling of chilliness.

Onset and course. The onset was about eight years before (in 1906) with an attack of headache and vomiting. The headache was very severe and constant, lasting about a month. Two months later he developed great thirst and entered a hospital in Toronto. He stated that he drank 10 to 12 gallons of water a day and passed 360 to 400 ounces of urine. After receiving medical treatment for about six months, he came to Chicago to work as a tailor. Since then he has been drinking much water and passing large quantities of urine. Up to April, 1915, he felt fairly well and then began to have nocturnal incontinence. One month before admission (July, 1915) he noticed increased fatigue and sensations of extreme heat on the soles of his feet and over the buttocks. The same day he passed about a quart of blood while attempting to urinate. He has been extremely weak since then. Sexual desire has been absent for eight years.

Past History. Until the onset of the present complaint he had been in good health. He passed through an attack of smallpox when three years old which was complicated by an infection in the right eye and loss of vision in that eye. He had a chancre in 1901 and gonorrhoea three or four times.

Family History. He has one child, eleven years old, living and well. His wife is divorced.

Physical Examination. The patient was very anemic and appeared to be in a weakened condition. The right cornea was opaque and there was no vision in that eye. The teeth were carious and the mucous membranes pale. A few subcrepitant rales were found in the right side of the chest. There was almost total absence of pubic hairs and the testes were small and atrophic. The prostate gland was tender; the epididymis appeared to be normal. An attempted cystoscopic examination was unsuccessful because the urethra was too small to admit a catheter. Examination in the eye depart-

ment on August 27, 1915, showed normal vision in the left eye and total blindness in the right eye due to phthisis bulbi. The visual field showed no scotomata or hemianopia. The fundus was normal.

Laboratory Findings. Examination on August 24, 1915, gave the following data: Urine volume, 2,000 c.c.; specific gravity, 1.012; reaction alkaline, no albumin, no sugar; spinal fluid Wassermann, negative; hemoglobin, 70%; red blood cells, 3,040,000; color index, 0.9 plus; white blood cells, 10,500; differential count, 54% polymorphonuclear, 40% small mononuclear, 5% large mononuclear, 1% eosinophile.

A radiogram of the skull on August 19, 1915, disclosed a definitely abnormal outline of the sella. The posterior bony portions, including the posterior clinoid processes, were absorbed, indicating pressure by an enlarged pituitary gland.

The diagnosis made by Dr. Joseph L. Miller was diabetes insipidus, hypophyseal dystrophy and cerebral lues. The treatment consisted of mercury rubs, iodides by mouth and hypodermic injections of pituitrin three times daily. A sugar tolerance test was made by Dr. Russell Wilder, November 13, 1915, using intravenous injections of various strengths of sugar solutions by the Woodyatt method; this showed tolerance at the lower limit of normal, or 0.8 gm. per kilogram body weight per hour.

The patient was discharged at his own request on December 28, 1915, somewhat improved.

He was readmitted on June 19, 1916. His chief complaint at this time was inability to control his urine. Examination showed him to be well nourished, not acutely ill, or in discomfort. The skin was white and smooth over the entire body. There was a noticeable lack of hair over the chest and a scanty supply on the chin and face, and almost complete absence of hair on the shins and arms. The skin, mucous membranes and tongue were extremely dry. The systolic blood pressure was 135, diastolic 90, pulse pressure 45. The blood and spinal fluid Wassermann tests were negative. June 30, 1916, the average daily volume of urine was 3,000 to 3,500 c.c., and dropped to 2,000 to 1,800 c.c. with intramuscular injections of pituitrin. A renal function test yielded the following result: phenolsulphonphthalein first appeared in 25 minutes; 5% in the first hour and 10% in the second hour, making a total of 15% elimination of the dye in the first two hours.

The patient left the hospital July 21, 1916, and returned for the third time December 2, 1924. He was then 50 years old. In addition to previous symptoms there was nausea and severe vomiting and he had become so weak that he was unable to work.

Physical examination showed him to be very pale and poorly nourished. His blood pressure was up to 164 systolic and 98 diastolic, pulse pressure 69. The reflexes were exaggerated, there was

a bilateral positive Babinski sign and ankle clonus. The skin was lemon yellow in color, dry and harsh.

Laboratory Findings. The hemoglobin proved to be 60% of normal; red blood cells, 2,800,000; leucocytes, 9,800; polymorphonuclears, 58%; lymphocytes, 36%; large mononuclears, 6%. Urinary excretion ranged between 2,500 and 3,000 c.c.

A roentgenogram of the skull on December 5, 1924, showed the sella to be shallow in proportion to the antero-posterior diameter, being deeper in the dorsal than the anterior half; the floor was well defined.

The patient was discharged January 19, 1925, and returned for the last time March 21, 1925. He was then so weak he could not walk, had lost 30 pounds in a few months and frequently vomited a greenish material unrelated to food taking. Polydipsia and polyuria had disappeared for the past few months. Frontal headache had been present for a few days, but no convulsions.

Physical examination revealed a white male about 56 years old, poorly nourished, with a lemon yellow tint to the skin and apparently acutely ill. His pulse was of very poor quality and thready. He was mentally sluggish. Blood examinations showed hemoglobin, 65%; red blood cells, 3,230,000; white cells, 24,200; polymorphonuclears, 85%; lymphocytes, 15%; no abnormal red cells were noted. The urine was reduced to 450 c.c., with specific gravity, 1,008; no albumin and no sugar. The temperature was normal or subnormal throughout. He continued to vomit excessively, being unable to retain anything by mouth, and died April 1, 1925.

SUMMARY OF THE CLINICAL HISTORY

The history is that of diabetes insipidus of about 20 years' duration, the onset occurring about 5 years after a primary lesion, with symptoms suggestive of syphilitic meningitis. At first the polydipsia was extreme and the polyuria enormous. Both improved temporarily under anti-luetic treatment and were partially controlled by hypodermic injections of pituitrin. Signs of hypophyseal dystrophy developed, consisting of loss of body hair, moderate adiposity and atrophy of the genitals, constituting the adult type of Froelich's syndrone or dystrophia adiposo-genitalis. During the last few months before death his diabetes insipidus had apparently disappeared.

AUTOPSY FINDINGS

The autopsy was performed by Dr. H. Gideon Wells on the day of death.

External Appearance. The body was that of a slender, poorly nourished but not emaciated man, appearing much younger than his

age. The hair of the head was abundant, dark, and with a few gray hairs. Eyebrows were present, but not heavy. The skin was pale. There was some blood crust on the lips and gums, and the mucous membranes were pale. The teeth were in bad condition. The sclerae appeared normal, but the right cornea was opaque. The superficial lymph nodes were not enlarged. There was a marked sparseness of body hair—none at all in the axilla, five or six in the pubic region, and about as many on the chin; none on the scrotum, thighs, arms, or elsewhere on the face or body. The testicles were small and soft. The penis was normal. There was a suggestion of superficial atrophic scars on the body of the penis. There was a slight palpable edema of both lower extremities and of the right wrist. There was distinct hypostatic edema. On opening the body, an abundance of subcutaneous fat of a lemon-yellow color, about 1 cm. in thickness, was exposed along the mid-line, not only over the abdomen, but also over the chest, and much more than would be expected from the slenderness of the body, that is, the subcutaneous fat resembled in distribution that of the female rather than the male, and there was a distinct thickening of the fat above the pubis to form a definite mons veneris.

Mouth and Pharynx. The tongue showed atrophy of the dorsal surface. The central part was covered with blood-stained crusts. The tonsils were atrophic, but contained pockets with a cheesy content. The lymphoid apparatus at the base of the tongue was distinctly atrophic.

Thyroid and Thymus. The thyroid gland weighed about 10 gms. The parathyroid glands could not be definitely located. The thymic body was replaced by fatty adipose tissue.

Heart, Aorta and Vessels. The heart appeared about normal. There were yellow patches in the aorta, especially in the arch. The aorta was of about normal size and still elastic. It showed distinct areas of thickening and wrinkling at the orifices of the subclavian and innominate arteries. There was a depressed calcified area, 10x15 mm. in the arch. The orifices of the coronary artery were of normal size, but there was a definite sclerotic patch in the anterior coronary artery just beyond the orifice. The posterior coronary artery also showed patches of sclerosis.

Liver. The external surface of the liver showed numerous depressed scars. It weighed 960 gms. and was brown in color. The surface scars ran into the substance of the organ, from 5 to 15 mm. Cut surface showed no further scarring or other changes.

The pancreas was small and weighed 50 gms. It showed no gross abnormalities.

Adrenals. The left adrenal was very small and atrophic. The right resembled the left. The cortex was reduced to a mere line of yellow, the medulla showing less reduction, but all told each adrenal was scarcely a millimeter thick, and about half the usual length and

breadth. They showed central postmortem softening and were so much involved in the perirenal fibrosis that they could not be successfully isolated for weighing.

Kidneys. The kidneys were excessively adherent to the perirenal fat. The left kidney was partly converted into a sac by distension of the pelvis. The cortex was thin, 1-2 mm., and the pyramids were about 5 mm. wide. The capsule was adherent, but when torn showed a very granular surface. The right kidney resembled the left. No evidence of acute inflammation was noted.

Urinary Tract. The urinary bladder and ureters were greatly distended. The bladder reached to the pubis and showed marked hypertrophy of the wall, but the mucosa was not inflamed. The ureters were about 10-15 mm. in diameter, but not inflamed.

Generative Organs. The testicles were about equal in size and measured each about 10x15 mm. They consisted of pale homogeneous tissue with no visible glandular elements. The prostate was atrophied to such a degree that it was not readily discerned. The seminal vesicles were empty and atrophied to thin fibrous cords with brown centers. The urethra showed a small amount of scarring at the junction of the prostatic and penile portions, but was not completely occluded.

Brain and Meninges. The calvarium was thin and weighed 350 gms. It was symmetrical and showed nothing abnormal. It did not adhere unduly to the dura. The dura was thick and opaque. The amount of cerebrospinal fluid was rather large, but not greatly excessive. The optic chiasma was free, but there were adhesions in the pia about the infundibulum and floor of the skull, which adhesions seemed limited to this area. The brain as a whole showed a slight degree of atrophy of the convolutions, i. e., the sulci were increased in prominence. The brain weighed 1,440 gms. The pia and arachnoid showed no thickening indicative of a syphilitic leptomeningitis. The arteries at the base of the brain were not apparently thickened. The sella turcica was shallow and flat. The hypophysis was extremely small and reduced to a flat disc 11 mm. transversely, 7 mm. vertically, and 1-2 mm. thick, including the thickened capsule which was not to be differentiated from the thickened dura, with which it merged. There was no distinct hypophysis and its location was determined solely by the entrance of the infundibulum into the thickened fibrous tissue.

MICROSCOPIC EXAMINATION

The hypophysis, including the adherent dura and blood vessels, was imbedded in celloidin and the entire mass completely sectioned. The infundibulum, posterior lobe and pars intermedia were replaced by dense avascular scar tissue. No remains of the anterior lobe were found until the middle of the block, where there appeared a mass of glandular tissue measuring about 2 mm. in diameter. This

again gradually faded out into fibrous tissue. The cords of epithelial cells were compressed and the sinusoids dilated and filled with blood, giving the impression of passive congestion. A number of sections contain the carotid artery lying just outside the anterior lobe. This shows a moderate endarteritis and a small area of calcification in the media.

The base of the brain was divided into four blocks, imbedded in paraffin and cut in serial sections. These were stained with hematoxyalin and eosin, van Giesen and Mallory's connective tissue stains. The collections of ganglion cells scattered through the hypothalamic region and in the tuber cinereum appeared well preserved. The ependyma lining the third ventricle was intact. Nowhere was there any perivascular round cell infiltration. Some of the small blood vessels were slightly thickened, but no distinct endarteritis was present. Dr. Hassin examined these sections and stated that, in his opinion, the normal architecture of this portion of the brain was intact.

Kidneys. The capsule is thickened and adherent to the surface of the kidney where there is a layer of round celled infiltration replacing the outer part of the cortex and running along fibrous areas into the pyramids. There is a similar round cell area and a heavy fibrous layer beneath the epithelial surface of the pelvis. There remains but a small amount of renal tissue in which the tubules are dilated and filled with protein. The vessels show marked arteriosclerosis. No acute suppurative changes are present.

Testicles. The glandular elements are almost completely replaced by fibrous tissue. Remains of the tubules are seen, but none contain distinct epithelial structures.

The seminal vesicle is distinctly atrophic and collapsed.

Prostate. This consists of fibro-muscular tissue with a larger proportion of fibrous than muscular elements. There are a few collections of small collapsed tubules staining almost black with hematoxyalin as if partially calcified.

Adrenals. The cortex is extremely thin, the cells small and poor in lipoid. The medulla is abundant, and seems normal.

Thyroid. The acini are extremely small, although most of them contain colloid. The arteries are calcified. No other changes are seen.

Thymus. The thymus shows numerous areas of lymphoid tissue scattered through the fat, slightly more than is usual at this age.

Pancreas. Seems entirely normal.

Liver. There are scars beneath the capsule. The lobules and cells are small, but no diffuse fibrosis or other changes were noted.

Aorta. The aorta was much thickened with atheroma in the intima and occasional old scars cutting across the media, but there are no active syphilitic changes.

Tongue. A section near the base shows an absence of lymphoid tissue, which is replaced by fibrous tissue.

Anatomic Diagnosis: Bilateral hydro-uretero nephrosis with chronic interstitial nephritis; hypertrophy and dilatation of the urinary bladder; syphilitic aortitis; slight aortic endocarditis; syphilitic scars of the liver; marked atrophy of the testicles, prostate, seminal vesicles, adrenals, thyroid and hypophysis; feminine distribution of body fat; nearly complete absence of body and facial hair; meningeal adhesions about the infundibulum; bilateral adhesive fibrous pleuritis; multiple healed nodular tuberculosis of both lungs; fibrous prostatitis; diffuse suppurative bronchitis and tracheitis; left suppurative otitis media and mastoiditis; atrophic glossitis; slight hypostatic subcutaneous edema; opacity of the right cornea; slight general atrophy of the viscera; atrophic pulmonary emphysema; slight brown atrophy of the heart and liver; incomplete stricture of the urethra.

SUMMARY OF THE PATHOLOGIC FINDINGS

How do the pathologic findings account for the clinical picture? The diabetes insipidus must be attributed to destruction of the posterior lobe of the hypophysis. The tuber cinereum and the entire base of the brain were normal in architecture and no grossly recognizable lesion was found in sections stained by the ordinary methods in use. The polyuria disappeared because of renal insufficiency, the terminal symptoms being those of uremia, resulting from stricture of the urethra, repeated ascending urinary infections and chronic interstitial nephritis. The ultimate cause of death was a complication of gonorrhoea, and his first illness was due to syphilitic basal meningitis with possibly a gumma in the posterior lobe of the hypophysis.

The polyglandular syndrome is not easily explained. The same difference of opinion exists regarding the basic pathology of Froehlich's syndrome as is the case with diabetes insipidus. I am inclined to believe that there was enough reduction in anterior lobe substance to produce functional insufficiency. On the other hand, syphilis may have produced diffuse atrophy of the testes, and the changes in the other endocrine glands occurred secondary to this. Yet diffuse atrophy of the testes is common in syphilis of long standing, and it is usually not accompanied by changes in other glands of internal secretion.

A number of points should be emphasized. First, that in our conception of the pathology of diabetes insipidus we cannot ignore those cases in which the lesion is confined to the posterior lobe and infundibulum of the hypophysis, nor the fact that when polyuria develops in a case of malignancy, it is

almost a pathognomonic sign of the presence of a metastasis in the posterior lobe of the hypophysis. Second, that the hypophysis and midbrain are anatomically united and in view of our present knowledge we must consider the hypophysis and the centers in the midbrain as a functionally united system. The rhythmic automaticity of the respiratory centers is dependent upon the chemical composition of the blood, and we may logically assume that the regulatory activity of the vegetative centers in the midbrain is influenced by the internal secretory function of the hypophysis. This is the view recently expressed by Berblinger (9), and is consistent both with the facts obtained from a study of pathologic anatomy, including circumscribed lesions of the hypophysis that cannot be duplicated artificially, and the evidence of experimental physiology and anatomy. The purely nervous theory of the genesis of polyuria no more solves the problem of diabetes insipidus today than did Claude Bernard's demonstration of piqure centers in the floor of the fourth ventricle solve the problems of both diabetes insipidus and diabetes mellitus.

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THE THERAPEUTIC INDICATION FOR THE USE OF ANTERIOR PITUITARY LOBE SUBSTANCE.*

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The practice of organotherapy is not a modern idea. It dates back many centuries when medicine was shrouded with mysticism and superstition. Progress was necessarily slow, as it had to advance along empirical lines. Although Hippocrates, Celsus and Dioscorides employed certain animal tissues in the treatment of diseases and the four London Pharmocopeias of 1618, 1650, 1677 and 1721 abounded with such remedies as blood, bile, bones, brains, excrement, fat, hearts, marrow, urine, etc., they finally fell into disrepute largely through Thomas Sydenham, who did not attach much importance to them, and later through William Heberden, who made a critical onslaught on polypharmacy and banished these remedies from the British Pharmacopeia in 1788. From this period until quite recently, when the modern sciences were beginning to be applied to medicine, little effort was made to re-introduce them. However, owing to a more comprehensive knowledge of the anatomy, histology and physiology of the glands of internal secretions and the various symptomatic manifestations produced through the arrest or alteration of their secreting function by modern scientific methods of experimental research on animals, definite clinical, pathological and anatomical bases were established for organotherapeutics. It is surprising to note the rapidity with which the indications have been met and how closely therapeutics followed the wake of physiological investigation. This has been particularly true of the thyroid, which served as the entrance wedge into the domain of endocrinology. Advancement in the study of the hypophysis was relatively slow because of the inaccessibility of the gland. It was not until 1906, when Paulesco developed a special surgical technique that a portion of the gland could be extirpated without death resulting to the animal. As a consequence of innumerable subsequent experiments the function of the hypophysis is now pretty well understood. Briefly these experimental results may be summarized as follows:

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A. The removal of the whole gland is uniformly fatal, producing tremor, muscular fibrillation, low temperature, slow pulse and respiratory rate, stupor and coma.

B. Removal of the posterior lobe does not cause death, although a deficiency causes hypotension, increased sugar tolerance, decreased basal metabolism and asthenia.

C. Complete removal of the anterior lobe causes a cachexia which terminates in death.

D. Partial removal of the anterior lobe causes obesity, genital hypoplasia, faulty skeletal development and hypothermia; the latter can be prevented by injection of an extract of the anterior lobe. In as much as organotherapy can only be rationally applied to deficiency glandular syndromes, the effects produced by overfunctioning of the pituitary gland need not be considered here.

Reasoning on the basis of cause and effect, it would appear that the administration of anterior pituitary lobe extract should yield favorable results in conditions arising from insufficiency of the lobe, namely, retarded skeletal growth, pituitary obesity, delayed puberty, genital hypoplasia, and hypophyseal cachexia.

Unfortunately the pharmacology of the anterior lobe is still meager as compared with that of the posterior lobe. In 1916 Robertson obtained a soluble lipoid, which represented some of the active principles of the anterior lobe and exerted a control over growth; to this substance he applied the name tethelin. Various feeding experiments of anterior lobe on animals and tadpoles by numerous investigators led to a division of opinion as to its pharmacodynamic effects. These discrepencies were apparently due to the age, species and growth period of the animals, the amount of gland feeding and the duration. The final analysis, which conclusively showed that anterior lobe administered orally has a distinct therapeutic effect in deficiency syndromes, depended upon the results obtained by the extensive use of this substance in the human being, so that its action is no longer a mooted question. The important desideratum is the correctness of the diagnosis in order to ascertain the proper indication for its use.

My personal interest in anterior pituitary therapy was incited by the study and observation of a patient who came under my care in 1915. This case, in addition to a group treated subsequently, was reported under the caption of "Fat Re-distribution in Dystrophia Adiposogenitals," in the Journal of American Medical Sciences, November, 1918, p. 714. The case referred to was a woman aged 41, married and weighing 125 pounds. She was a confirmed invalid by reason of three fairly well defined groups of signs and symptoms, namely: those referable to pituitary insufficiency; girdle adiposity and genital hypoplasia; those referable to gonadal insufficiency: frigidity, sterility and amenorrhoea, and those referable to thyroid insufficiency: apathy, stupor, dullness and neuromuscular and trophic skin disturbances. The outstanding feature was her obesity, although she had gained about twenty pounds in the previous ten months the only noticeable increase was confined to her hips, thighs, gluteals and lower abdomen.

Regarding her symptoms of pluriglandular origin in which the thyroid, pituitary and ovaries were involved, the three respective hormones were administered with singularly marvelous results. Her general condition rapidly improved so that she was able to resume her housework, but more striking was the change in her body configuration. Measurements obtained from her dressmaker, taken two weeks before treatment was begun and eleven weeks afterwards, showed a reduction in the size of her hips seven and a half inches and of the waist five inches. She had lost only one pound in weight. The face, neck, chest and arms, which before treatment presented the appearance of emaciation, had filled out and the configuration of the body returned to normal. Treatment was discontinued after five months and there has been no return of her symptoms.

This case afforded convincing proof that the therapeutic results in the girdle type of dystrophia adiposogenitalis cannot be gauged by the effect of glandular treatment upon the weight of the patient, and established the fact that a better criterion is a record of systematic circumference measurements which have been adopted. These measurements are the maximum hip, minimum waist, axillary, neck, thighs at the groin, knees and ankles. In the "System of Endocrinology and Metabolism," published

in 1922, the writer reported forty-six cases of pituitary fat dystrophy studied and treated in this manner. These patients usually received thyroid in doses of one to two grains a day and anterior pituitary lobe in doses of five to ten grains a day, although in some of the cases good results were obtained with anterior pituitary lobe alone.

The reason for administering thyroid in conjunction with anterior pituitary lobe is on account of its influence in accelerating general metabolism and its stimulating effect upon the pituitary and sex glands. Moreover, the thyroid gland is so closely related functionally with the pituitary that it is frequently a concomitant etiological factor. In this class of cases it has a dual action, namely: supplying the lack of thyroid hormone in an associated thyroid deficiency and stimulating the function of the anterior lobe of the pituitary. The tabulated results of the forty-six cases reported showed definitely marked improvement in twenty-six cases, in sixteen of which there occurred typical fat re-distribution as indicated by an increase of the circumference measurements above the waist and a decrease below the waist in addition to the general improvement of the patient. Nine cases showed moderate improvement, but without typical re-distribution of fat. Eleven cases remained unimproved. In most of these cases the treatment was not administered a sufficient length of time to derive any real benefit. Since the report of this series the number of cases treated has almost doubled with practically the same results.

In the Fröhlich type of dystropsia adiposogenitalis (or the pre-adolescent form) advice is often sought in regard to some of the cardinal features of the disease, such as stunted growth, sexual infantilism, delayed puberty, amenorrhoea, etc. If these conditions are caused primarily by reason of insufficient anterior pituitary secretion good results are obtained by pituitary feeding either by itself or in combination with thyroid.

In pituitary dwarfism in which skeletal growth was arrested at an earlier age than normal puberty and the epiphyses are still ununited, anterior pituitary feeding will frequently cause a renewal of skeletal growth even after cessation for a number of years. The following case is cited as an example: W. F., male, aged seventeen years and one month, weighed 51½ pounds and

measured 130 cm. in height, which; according to normal standards of height and body weight, represents the average boy eight years of age. He was normal at birth and developed normally, both mentally and physically, until this age, when he ceased to grow, although he continued to develop mentally. Besides cessation of growth he exhibited certain features characteristic of hypopituitarism, namely: maxillary prognathism; absence of beard, hirci and crines pubes; soft, delicate skin; infantile voice; genital hypoplasia; delayed epiphyseal union and sellar deformity. Treatment was begun in October, 1921. In this case whole pituitary gland was administered in doses varying from two to six grains a day, with the hope of benefiting the asthma from which he was suffering as well as to promote skeletal and genital growth. In December he measured 131½ cm., February, 1922, 132¾ cm., and September, 1922, 137 cm., which represented a gain of 7 cm., or almost three inches, in less than a year. Besides, he gained twelve pounds in weight. He was relieved of asthma and felt perfectly well. No thyroid was administered.

In other cases the same effect upon growth was produced by feeding anterior lobe alone. The reason for citing this case is to demonstrate the fact that certain cases of asthma associated with disturbed function of the pituitary apparently yield to pituitary therapy which is attributed to the effect of the posterior lobe. Unquestionably the stimulating effect upon growth is due to the anterior lobe extract. Crofton (An Outline on Endocrinology) reports a boy aged seventeen years who grew five inches in less than a year on two grain doses of anterior pituitary lobe. McGraw observed that in cretinoid dwarfs, growth is much accelerated by administering anterior pituitary lobe extract in addition to thyroid.

Delayed puberty: By the experimental feeding of rats with anterior pituitary lobe, some observers have shown that the animals matured sexually much earlier than their controls—according to Goetsch, in one-third of the normal time. Posterior lobe extract produces no such effect, in fact it exerts an inhibiting influence upon growth. The stimulating effect of anterior pituitary lobe upon growth applies to both the genital system and to growth in general. One can thus appreciate the rationale of

administering anterior pituitary lobe in delayed puberty and sexual infantilism in early life and distorted sexual functions such as loss of libido and potentia in adult males and amenorrhoea, frigidity and sterility in adult females, especially when these conditions are associated with other signs and symptoms of hypopituitarism. Splendid results have been obtained in boys in whom the secondary sex characteristics failed to appear at the age of puberty and likewise good results have frequently been obtained in girls who did not menstruate at the age of fifteen or sixteen. In this group of cases the treatment is more efficacious if small doses of thyroid are added.

The following case illustrates the effect of organotherapy upon amenorrhoea in a woman with adult hypopituitarism. She was twenty-eight years old and married, but had no children. Following an attack of influenza she developed a state of hypopituitarism. Her weight increased from 158 pounds to 228 pounds in four years. At examination she presented the following features: marked girdle adiposity, soft delicate skin, macroglossia, hypotension, increased sugar tolerance, monomucleosis, abnormally large open sella, bitemporal headaches, amenorrhoea and sterility. A diagnosis of hypoplasia of the reproductive organs was made by Dr. Richardson. Menstruation ceased in September, 1922, and had not returned at the time of examination, July 21, 1923. At her last consultation, January 28, 1924, records show that she menstruated in October, December and January. She had lost fifty-one pounds and her body configuration was almost normal. The headaches had disappeared and, according to her own statement, her strength and endurance had increased 100 per cent. The treatment consisted of two grains thyroid and five to ten grains anterior pituitary lobe daily.

The epileptic episodes which occasionally accompany pituitary disorders are sometimes favorably influenced by organotherapy. Cushing reported several cases in which pituitary feeding ameliorated the attacks, and Beverly Tucker reported a series of cases in which it was of marked benefit. Other observers have reported cases apparently cured by this treatment. Pituitary was administered either in the form of whole gland or anterior lobe extract. The favorable results obtained in a

patient under my personal supervision have been attributed to organotherapy. The patient referred to me by Dr. Maxson was a boy aged 13 years with a negative family history. He was normal at birth and his physical development was normal as a child. At an early age he manifested a nervous trend. He was restless and emotional and cried almost daily. At six he entered school. He was mentally delinquent and unable to keep up with his studies and make his grades in school. At eleven years of age he had an attack of influenza, which was followed by epileptic seizures. The severe attacks were characterized by sudden onset with fall, loss of consciousness and tonic and clinic spasms. In the milder forms the loss of consciousness was not complete and the spasmodic contractions were slight. The seizures increased in frequency, occurring almost daily; sometimes he had as many as four or five in one day. On May the tenth, 1920, treatment was begun with anterior pituitary lobe and small doses of thyroid. The seizures promptly ceased. On July 26th, 1920, his pastor made the following statement in a letter,—"he is doing fine. He has not yet suffered any check in progress and bids fair to become a real boy, just as surely as he bade fair to become an imbecile." The fact is that he became mentally alert, pursued his studies and passed his grades creditably through high school. He has not had any attacks now for a period of six years and at present is actively engaged as a mechanic in a garage where he measures up to the standard of a normal healthy individual. In this case the mental processes were undoubtedly accelerated by treatment and the delinquency, which was an important factor, was completely overcome. Other instances have occurred in my practice in which marked improvement was noted in this regard. Only two weeks ago a girl with pituitary obesity and mental sluggishness, which retarded her progress in school, volunteered the information that she is doing much better in her class work and that her examination marks in two branches were one hundred. This improvement occurred while she was taking anterior pituitary lobe. It is doubtful whether it is of any value in psychiatric practice, although some psychiatrists state that improvement followed its administration in certain forms of psychoses.

General consideration: From the clinical results obtained by the oral administration of anterior pituitary lobe in carefully

selected cases over a period of ten years the writer is convinced of its merits as a therapeutic agent. During the same time he has learned to recognize its limitations. After all, the indications are few and can be established only by a complete and thorough study of the patient in reference to pituitary function as well as the function of other endocrine glands which may be simultaneously affected. The hormopoetic system with its chain of glands is analogous to the hemopoetic system which comprises the circulating blood, spleen, bone marrow, lymphatic glands, etc.; any one, two or more of these organs may be concerned etiologically in diseases of the blood, the picture being dominated by the organs involved. The same is true in diseases of the ductless glands, which may be uniglandular but are more frequently of pluriglandular origin. The relative importance of the respective glands involved must be evaluated by a careful analysis of the history and clinical manifestations before considering glandular therapy. Whether one is justified in feeding mixed gland depends upon such an analysis. Failure in obtaining satisfactory results are often due to injudicious combinations or over-doses.

In order to obtain the best result with anterior pituitary lobe it is necessary that the gland preparations be fresh, the dose adequate but not excessive and the duration of treatment extended over a sufficient period of time—rarely less than six months, frequently a year or more. In a large number of cases treated, no advantage was seen in giving large doses from fifteen to thirty grains a day over smaller doses of from five to ten grains a day. On account of the protracted period of treatment, additional expense and discomfort to the patient, the hypodermic method was found impractical. Moreover, there was a greater tendency for patients to abandon treatment long before the desired results were achieved than in those to whom treatment was given orally.

Whenever thyroid was administered in addition to anterior pituitary lobe its tolerance was first established by beginning with small doses, one-quarter of a grain to one-half of a grain twice a day and increasing gradually over a period of two weeks before anterior pituitary lobe was added. The maximum dose of thyroid was rarely over two or three grains daily.

Abstract Department

Intracardiac injection of epinephrine in apparent death of new-born. Garipuy & Mériel (P.), Presse méd. (Par.), 1926, **34**, 180-181; Abst., J. Am. M. Ass., **86**, 1227.

Reanimation of the heart by intracardiac injection of epinephrine is being more and more frequently reported by reason of the simplicity of the technic. Especially in syncope resulting from chloroform, the method has proved good. It was logical, therefore, to employ it in other cases of arrested heart beat. Garipuy and Mériel of Toulouse report a favorable result from intracardiac injection of epinephrine in a case of asphyxia in the new-born. In their case of an infant born apparently dead, without heart beats or pulsation of the umbilical cord, aspiration of mucus and artificial respirations had proved ineffectual. After five minutes an intracardiac injection of 1 cc. of a 1:1,000 solution of epinephrine was administered in the fifth left intercostal space midway between the median and mammillary lines. Almost immediately the infant heaved a sigh and the heart began to beat. Artificial respiration was continued, and at the end of fifteen minutes normal respiration was established. The authors pointed out that, in infants, intracardiac injection is exceedingly simple. The lungs of the infant that has not yet breathed lie flat against the vertebral column. There is, therefore, no danger of injuring them in making a puncture between the median and the mammillary lines. After the lapse of from ten to fifteen minutes, all hope of resuscitation will be gone. Therefore, no time should be lost, and, when ordinary maneuvers have failed, intracardiac injection of epinephrine should be given without delay. It will have every chance of being effectual if it is done within five or six minutes after the arrest of the heart beat. After the injection, artificial respiration and rhythmic traction of the tongue should be kept up. The injection of epinephrine is not supposed to supplant, only to supplement, these methods.

Epinephrine injections in relapsing fever and malaria. Mishtchenko (I.), Prophylaktitcheskaia Medizyna (Kharkoff), 1926, **5**, 20-24; Abst., J. Am. M. Ass., **86**, 1408.

Mishtchenko recalls Zlatogoroff's epinephrine test in relapsing fever. A dose of 0.5 cc. of 1:1,000 epinephrine solution is injected subcutaneously in patients with relapsing fever, and the temperature is taken every two hours. If the temperature does not rise, the

injection is repeated 48 hours later. If the temperature continues within normal range after the second injection, this is accepted as a negative reaction. With a positive reaction, fever occurs within twelve hours after the injection. Personal experience has confirmed that relapses need not be feared as long as the epinephrine test is negative. The positive test was most pronounced between the fourth and seventh days following an attack. Contraction of the spleen and consequent expelling of specific spirochetes into the general circulation may explain the positive response to the epinephrine. The test was always negative in cases of reinfection. The epinephrine test was valuable also in diagnosis of malaria treated recently or insufficiently. The mechanism was the same, the expelling of plasmodia into the blood. The injection seemed harmless.

Adrenal and nervous factors in blood pressure (Intervention des réactions adrénalino-sécrétoires neuro-vasculaires dans la correction des troubles de la pression artérielle chez le chien). Tournade & Chabrol, Société de Biologie, 1926 (April 24); Abst., Paris M. J., **16,** 475.

The authors have shown that the lowered pressure resulting from the loss of a large amount of blood from the carotid artery in the dog provokes simultaneously: (1) a discharge of adrenalin as shown by the method of anastomosing the adrenal vein of one animal to the jugular of another; (2) a vasoconstriction of the kidney, resulting in change of volume of this organ, previously lowered by an irrigation of blood at constant pressure. This is done by direct perfusion, through anastomosis of the arterial circulation of a normal dog with the renal artery of the experimental dog.

—R. G. H.

Adrenals and oestrous cycle (Surrenale et cycle sexual). Watrin (J.), Rev. franç. d'Endocrinol. (Par.), 1926, **4,** 45.

The author proposed to verify with guinea pigs the experiments which Riddle made with pigeons. He injected young immature guinea pigs with varying doses of from 2 to 4 cc. of the follicular fluid obtained from puncture of Graafian follicules that had reached maturity. The modifications noted were intense congestion of the vagina, uterus and tubes. With respect to the adrenals, however, he observed nothing except a slight congestion of the cortical substance. He concludes that the follicular phase of the oestrous cycle does not influence the adrenal glands, at least in guinea pigs, and he believes this is also true in rabbits because he has never observed any reaction in the adrenals of rabbits when the Graafian follicules were mature.—R. G. H.

New clinical aspects of the Steinach operation. Benjamin (H.), M. J. & Record (N. Y.), 1925. Reprint pp. 1-39.

The author reports upon his results with the Steinach operation in 114 cases. A considerable number of case studies are included. He concludes that the principal indication for the Steinach operation is premature or physiological old age. In 77% of the cases it has been more or less definite, sometimes striking results in alleviating symptoms of senility, thus prolonging the active period of life. These results can be obtained only if the correct technic is used and a definite indication insisted upon, and under these conditions the procedure is definitely harmless. Where no reason exists to preserve the procreative ability of the patient, a bilateral operation is preferable. The results last several years. Functional impotence is only an indication: (a) when it is one of the symptoms of senility (then senility is the real indication; (b) when endocrine gonadal deficiency can be assumed as a cause; (c) as a last resort after all other treatments have failed.—R. G. H.

Vasectomy in a dog (Un interessant resultat de vasectomie chez un chien). Berganer (V.), Comp. rend. soc. de biol. (Par.), 1925, **93**, 1575-1576.

Sectioning of the left deferent canal in a 14-year-old dog was followed in 4 weeks by disappearance of a central cataract and of a cough, improvement in sight and general physical condition and increase in weight and activity. Four months later the animal died of pulmonary edema. The pH of the blood and stability of the serum colloids (Ruzicka-alcohol precipitation method) were determined throughout the experiment. There was no appreciable change in pH. An increase in the amount of alcohol required to produce cloudiness indicates an increased stability of the colloids which the author interprets as due to increase in metabolism following the activation of the pubertal gland.—R. R. Durant.

Interstitial gland cells in the human ovary. Lewin (B. D.), Am. J. Med. Sc. (Phila.), 1926, **171**, 518-520.

Among 43 pairs of ovaries from psychotic patients, 2 ovaries, each from a different case, showed in the hilum a small island of cells identical in appearance with the interstitial gland cells of the testis. The clinical diagnosis in both cases was "senile psychosis." No functional features were to be correlated with this finding. If the cells exerted "male" influences, the fewness of their number in the presence of general senility, prevented this influence from manifesting itself. The possibility of overlooking the presence of such an island, when the ovary is removed in the usual routine autopsy manner, is pointed out.—R. G. H.

The female sexual hormone. III. The effect of the cyclic hormone when administered by mouth. Loewe (S.), Lange (F). & Faure (W.), Deutsch. m. Wchnschr. (Berl.), 1926, **52**, 310-313; Abst., Chem. Absts., **20**, 1440.

With mice the authors were able to obtain the typical specific effects of the sexual hormone when the oral method was used; the dose required was 20 times the subcutaneous dose. Fractional doses under certain conditions produced an additive effect.

Specific inhibitory hormone of corpus luteum. Papanicolaou (G. N.), J. Am. M. Ass. (Chicago), 1926, **86**, 1422-1424; Abst., A. M. A.

In experiments performed on guinea pigs, the author found that after complete removal of the corpora lutea the ovulation and estrus return on the eleventh day, while in normal guinea pigs estrus usually returns at the fifteenth or the sixteenth day. These findings indicate that the corpora lutea exert an inhibitory action, suppressing and delaying the process of ovulation in the rhythmic function of the ovary. The author has used this inhibitory function of the corpus luteum as a criterion for studying luteal extracts and especially for the recognition of the specific luteal hormone. Some of the purified extracts have been highly efficient in their inhibitory effect. An unmistakable delay follows each injection of this hormone given in small quantities of from 1 to 2 cc. By giving weekly one injection of from 1 to 2 cc., it is possible to suppress the process of ovulation indefinitely. Through such a procedure, Papanicolaou has been able to suppress ovarian rhythm in several animals for a period of from 2 to 4 months. At the end of this period the injections were discontinued, and ovulation reappeared several days after the last injection. The ovaries of animals killed after long suppression show a complete absence of corpora lutea and a large number of middle sized follicles with a highly congested theca interna. This congestion of the theca interna causes an early atresia and prevents the follicles from enlarging into mature graafian follicles. The old corpora lutea degenerate gradually, and the ovary becomes practically free of luteal tissue. The full growth of the follicles and the process of ovulation are thus suppressed, and the ovary thereby loses its cyclic rhythm. This effect is evidently specific and is caused only by the subcutaneous administration of luteal extracts. Similar extracts prepared from other parts of the ovary (follicular fluid, cystic fluid, ovarian residue) and from other organs (placenta, thyroid, thymus, testes and immature ovaries) in all cases gave negative results. It seems evident that this particular hormone is typical for the corpora lutea.

Eunochoid adiposo-genital dystrophy (Sur la Clinique et le Traitment de la Dystrophie Adiposo-Genitale Eunuchoidique). Schere-

schewsky (M.), Rev. franç. d'Endocrinol. (Par.), 1925, **6**, 395-407.

The author reports in detail a case of dystrophia adiposogenitalis. By way of treatment a cryptorchid testis of a healthy man was transplanted subperitoneally. Histologically the graft showed deficiency of spermatogenic tissue, but augmentation of interstitial cells. The therapeutic results were nil.—R. G. H.

Activation of the sexual cycle, development of sex characters, reactivating effect on the senile female organism by ovarian and placenta extract. Steinach (E.), Heinlein (H.) & Wiesner (B. P.), Arch. f. d. ges. Physiol. (Berl.), 1925, **210**, 598-611.

Steinach, in conjunction with his associates, Heinlein and Wiesner, have during the last two years conducted a series of experiments on rats and guinea pigs, with a new stable extract prepared from the ovary and the placenta. It was found that in infantile castrates that had not shown any cycle, two to four injections given every 24 hours produced the onset and normal course of a cycle, which could be reproduced by repeated injections ad libitum. Every experiment was positive. A cycle was shown and proved by daily microscopic examination of the vaginal secretions. The size of the uterus developed in the injected infantile castrates exactly the same as in normal animals. The otherwise atrophic state of the vagina was likewise counteracted and brought to full development. The article gives the exact protocols of the experimental animals and their controls. Castration of fully developed animals normally causes a definite cessation of the sexual cycle while the uterus gradually atrophies. With two to four injections given every 24 hours, animals castrated seven months before had their sexual cycle restored and uterus and vagina remained normal. The extract thus forms a potent substitute for the typical endocrine function of the ovary. In a normal female rat the sexual cycle ceases in the nineteenth to the twenty-second month. Animals were injected when the cycle had definitely ceased for two to five months. In every case the experiment was positive, the cycle reappeared and the senile-atrophic tissue of uterus and vagina was regenerated. For several months the sexual cycle recurred normally and automatically without further injections. Accompanying the restoration of the ovarian function, the restoration of the entire organism and the improvement of the animal's general condition was observed in a very pronounced way: the emaciated animal began to eat again, gain in weight, new hair grew on bald spots and the entire fur became thick and glossy. In albinos the iris distinctly showed better circulation. The senile attitude disappeared. The animals became vivacious, curious and began to clean themselves again. They regained the interest of the males who had entirely ignored them before and normal sexual intercourse was observed. A num-

ber of extract protocols are given. Infantile castrates treated with injections every 48 hours for three weeks showed normal mamillae compared to the atrophic, almost invisible infantile organs of the non-treated control animals. The extract had no apparent harmful influence either in castrated or in normal animals. Other ovarian and placenta extracts on the market were found utterly useless; likewise extracts from other organs, for instance, the liver, prepared according to the same method as the author's extract, had no effect whatever. It will be difficult to standardize the extracts, but it seems as if there is a certain relationship between the weight of the animals and the weight of the amount of extract injected. The extract produces exactly the same results as ovarian implantation. The work of other investigators, as well as Steinach's own experiments bearing on the present subject, are fully discussed in a supplementary chapter.—H. Benjamin.

New researches upon the injection of follicular liquid (Recherches nouvelles sur les injections de liquide folliculaire). Watrin (J.), Rev. franç. d'Endocrinol. (Par.), 1926, **4**, 48.

The injection into immature females of follicular liquid taken from a mature ovary causes intense congestion of the genital tract. The tests made by the author confirm this. The congestive phenomena, sometimes very marked, affect other viscera. They appear promptly and disappear rapidly after the cessation of the injections. Under the influence of these injections one is able to produce a rupture of the vessels of the uterine musculature. In a young girl with depressed menstruation an injection of the follicular liquid made 6 days before the expected appearance of normal menses was followed after 24 hours by the beginning of flow.

Studies in experimental diabetes insipidus. Bourquin (Helen), Am. J. Physiol. (Balt.), 1926, **76**, 181-182.

Diabetes insipidus has been produced in dogs by exposing the hypophysis and directing an electric cautery into the mammillary bodies. That the effective lesion is injury to the mammillary bodies has been demonstrated by histological examination of the brain stem of nine dogs sacrificed while diabetic and of four others, which were not diabetic, but injured in the region of the base of the hypophysis. The lesion common to all of the first group was injury of the mammillary bodies. In four that was the only lesion. The lesions in the latter group were in the tuber cinereum of two, lateral to the mammillary bodies of two. Dogs can be rendered diabetic repeatedly in this way. If, however, the lesion is severe a diabetes already established is immediately stopped and cannot be produced. Histological examination of the brain stem of eight animals subjected to this type of experiment showed complete destruction of the mammillary bodies. Diabetes insipidus is, therefore, an irrita-

tive and not a deficiency phenomenon. It is entirely independent of the hypophysis. A group of four dogs was hypophysectomized and typical diabetes produced. In a second group of four dogs hypophysectomy was successfully performed without causing diabetes insipidus. The cause of the polyuria is still undetermined. It is not a primary thirst phenomenon as some claim, for typical diabetes is produced in dogs deprived of water after the operation. It is not due to an increase in blood chlorides, as others claim, for in 14 of 16 dogs, in which chlorides were determined under standard conditions before and after diabetes insipidus was produced, the diabetic blood contained the same amount as did the normal blood.

Action of hypophyseal extracts upon intestinal peristalsis and constipation (Action des extraits post-hypophysis sur le peristaltisme intestinal et sur la constipation). Carnot (P.) & Terris (E.), Paris M. J., 1926, **16**, 333-337.

The peristaltic action of the extract from the posterior lobe of the hypophysis is shown clearly under examination with the fluoroscope. It is marked by colonic contractions and a series of copious evaculations, completely emptying the colon. This action may be graded by various ways of injection and various doses following clinical indications and contra-indications. By vein the most energetic, most vigorous but most violent results are secured. In cases of occlusion, of intestinal paresis and of dyschesia of the sphincter it is especially useful. It is not to be used with a hypersensitive or an organically diseased colon. Intra-muscular injection is more convenient and gives remarkable effects, but is less energetic. They usually are adequate in cases of chronic intestinal stasis. Subcutaneous injections are still less effective. Simple ingestion is most convenient and gives excellent results in habitual constipation. It reinforces intestinal tonus and permits re-education of the musculature of the terminal part of the intestine.—R. G. H.

The action of pituitary extract administered by the alimentary canal. Knaus (H. H.), Brit. M. J. (Lond.), 1926, **1**, 234-235.

In the investigation described cats were used as the experimental animals. The author states that when pituitary extract is administered at any part of the alimentary canal—mouth, stomach, small intestine, rectum—pressor action is never observed. When pituitary extract is introduced into the stomach or small intestine, it exerts no influence on uterine muscle. When the extract is administered by mouth, after a latent period of about eight minutes, it exerts a marked action on the uterus, increasing both the tone and automatic movements. The oxytocic substance is therefore absorbed. The rectum absorbs the oxytocic substance also, although absorption is probably not so complete as with administration by mouth.

Experimental studies on growth. XVIII. Further experiments on the influence of tethelin upon the growth and longevity of the white mouse. Robertson (T. B.) & Ray (L. A.), Australian J. Exper. Biol. & M. Sc. (Adelaide), 1925, **2**, 173-188.

The authors have devised a new technique for the comparison of curves of growth. This consists in plotting the average weights of the animals at different ages not as a series of points, but as a series of areas within which the true values are as likely as not to lie. These collectively constitute a ribbon-shaped area, of which the center at any age is the experimentally ascertained average weight and the vertical width is twice the probable error of the average. Any curve lying outside this area will then represent the growth of some distinctively different group of animals. If two areas overlap for a considerable proportion of their extent, then, during that period, they represent the growth of identical animals. This technique was applied to the further investigation of the effects of tethelin upon the growth and longevity of mice. Previous researches had shown that if tethelin be administered in dosage of 4 mg. per day by mouth, administration during a considerable proportion of the life of the animals leads to the production of small animals which are unusually heavy for their size, a condition reminiscent of acromegaly in human beings. The duration of life is greatly extended, namely, about 100 days, or between one-sixth and one-seventh of the average life-duration of normal animals. If, however, the administration be limited to 8 weeks, from the fifth to the thirteenth inclusive, so that it is discontinued before sexual maturity has been attained, then the growth of the animals, which is at first retarded, is subsequently much accelerated, so that a veritable gigantism is ultimately attained, the average treated animals exceeding the average normals by as much as 25% in weight. Life-duration is not increased. In the present experiments the effect of brief pre-adolescent administration of tethelin was again investigated, the dosage being 4 mg. per day, administered subcutaneously. The results confirm those previously obtained when the tethelin was administered by mouth, but the excess of weight attained was not so great, only 13% above the average, although it lay far outside the range of probable error. On the other hand, the life-duration was greatly increased, almost as much as in previous experiments in which administration was continuous. The effects of hypodermic administration for a brief pre-adolescent period are, therefore, of a character intermediate between those obtained in response to oral administration for a like period and those obtained in response to oral administration for a more prolonged period.—Author's Abst.

Diabetes insipidus (A Propos d'une Observation de Diabete Isipide). Sabrazés (J.), Rev. franç. d'Endocrinol. (Par.), 1926, **4**, 12-23.

The author presents in detail the case history of a subject of diabetes insipidus who did not react at all to whole gland hypophysis treatment by mouth. So far as urine output was concerned, injections of posterior lobe extract had little influence, but they did relieve the intolerable sense of thirst and led to a feeling of euphoria instead of the depressions to which the patient was subject.

—R. G. H.

Pituitary extract to initiate labor at term. Tholen (M. H. G. A.), Nederl. Tijdschr. v. Geneesk. (Haarlem), 1926, **1**, 526-530; Abst., J. Am. M. Ass., **86**, 1592.

Tholem gave an intramuscular injection of a small dose of pituitary extract to forty-eight women who were waiting for labor at term, usually after rupture of the bag of waters. Labor pains followed in from half an hour to an hour in 76% of the primiparas and 77% of the multiparas. In five cases requiring premature delivery, the pituitary treatment had no apparent effect. In those cases in which only false labor pains followed the pituitary extract, a sedative was injected at once. The pituitary extract was preceded by castor oil, and the injection was repeated once or twice. The fetal heart sounds were constantly supervised, and no untoward by-effects were observed in either mother or child.

Variations in the blood sugar content following the administration of insulin. John (H. J.), J. Lab. & Clin. M. (St. Louis), 1926, **11**, 548-560.

Observations on the change of the blood sugar level in diabetic patients following repeated doses of insulin given intravenously during the day, hourly (or more) fluctuations and the same routine on several subsequent days on the same patient. The results are shown in a series of graphic charts over periods of 7 to 21 hours. The cases studied were of varying severity of diabetes, and on a standard dietary routine of gm. 100-60-128-1800 (C-P-F-Cals) so that the only variable was the insulin which was given in uniform and also in varying doses, in one, two or more hours during the day. In one coma case 20 units of insulin was given hourly for 11 hours and bi-hourly for 10 hours more. There is no absolute regularity in the fall of blood sugar after insulin. The degree of drop varies in the same individual on subsequent days on an identical routine. The angle of the fall varies from day to day, or there may be even a rise of blood sugar after such a dose, due no doubt to the post-prandial rise of blood sugar not being adequately counteracted, though the general rule is a drop of blood sugar after insulin. When the insulin dosage was varied it did not necessarily follow that the larger the dose the greater the fall, for the reverse was often the case. Frequent small doses of insulin accomplish more than a single equivalent dose; such an apparently cumula-

tive effect is independent of the size of the doses. Several insulin reactions were recorded, these happening at no marked hypoglycemic, in fact some at a marked hyperglycemic, level. Large doses of insulin were given bi-hourly, 20 units throughout the day with a continued blood sugar level between 68 and 52 mg. per 100 cc. without producing any reaction.—Author's Abst.

The possible relationship between acromegaly and diabetes. John (H. J.), Arch. Int. Med. (Chicago), 1926, **37**, 489-511.

Experience with the two cases that are reported, in which diabetes was associated with acromegaly, and a study of the literature led John to conclude that, in general, hypopituitarism is accompanied by an increased carbohydrate tolerance and hyperpituitarism, by decreased tolerance, but this is not an absolute rule. In the majority of cases of so-called diabetes associated with acromegaly that have been reported in the literature, especially in older cases, the diagnosis of diabetes was made on the basis of urine examination only. Since glycosuria does not always indicate diabetes, this should be borne in mind in evaluating these reports. In most reported cases and in the two here reported, the onset of acromegaly preceded the onset of diabetes by many years. The treatment of diabetes associated with acromegaly does not differ from the treatment of the ordinary case, and the response of the former type of case does not differ from that of the ordinary cases. Patients in whom diabetes is associated with acromegaly if not controlled are as subject to acidosis and coma as the ordinary case. Hypersection of the posterior part of the pituitary gland seems to produce hyperglycemia and glycosuria. Whether this is the predisposing factor is not certainly established; the incidence of diabetes in acromegaly is rather high, but, whatever the primary factor, the metabolic disturbance is due to a decreased insulogenic secretion. It may be that the factor that produces hyperpituitarism may also have some influence in producing the decreased insulogenic function which brings about diabetes. Diabetes is always due to the same immediate factor, namely, a diminution of the insulogenic function and whether it is or is not associated with pituitary dysfunction, its course does not vary and the same treatment is indicated as in the ordinary case.—R. G. H.

Absorption and insulin (Zur Resorptionsbeschleunigung Durch Insulin). Koref (O.), & Mautner (H.), Klin. Wchnschr. (Berl.), 1926, **5**, 191.

These investigators found that the gastro-intestinal tracts of rats which had received insulin were empty, while those of controls were full when the animals had been fed water, milk or an 8% solution of magnesium sulphate. The animals to which insulin was administered were also more susceptible to poisoning by alcohol

than the controls. If curate was fed 2 hours after the injection of insulin the animal showed typical symptoms in about one-quarter of an hour.—E. Larson.

Note on a rapid and simple method of preparing a highly active pancreatic secretion solution. Luckhardt (A. B.), Barlow (O. W.) & Weaver (M.), Am. J. Physiol. (Balt.), 1926, **76**, 182.

The first 100 to 150 cm. of a dog's intestine are excised and the intraintestinal contents removed by passing a stream of tap water through the isolated loop. One hundred to 150 cc. of 0.2% to 0.4% HCl acid are now introduced into the loop and the ends closed with hemostats. The loop with acid contents is either incubated at body temperature or allowed to remain at room temperature for 10, 30 or 45 minutes. Ten minutes' extraction at room temperature usually suffices. At the end of that time the clear or straw colored content of the loop is filtered through cotton (or filter paper) to remove small flecks of desquamated mucosa. The filtrate is now ready for intravenous injection. On intravenous injection of 5 to 10 cc. (10 to 14 kgm. dog under barbital-sodium anesthesia) a copious secretion of pancreatic juice ensues comparable often in quantity to that prepared by the prolonged method of Bayliss and Starling. The earlier such extractions are carried out following excision of the loop of gut from the animal, the less vasodilatation is seen following intravenous injection. We have prepared some extracts with pronounced secretagogue action which were virtually devoid of vasodilatins. Both vasodilatin and secretion dialyze through collodin sacs. When kept in brown bottles under toluol in the ice box, the preparations made by the method just described seem to lose their secretagogue action more rapidly than those preparations made from the mucous membrane by the usual process of maceration, grinding with sand and boiling with 0.4% HCl. The vasodilatin action persists, however, indicating anew that the secretagogue action of pancreatic secretin solution is not due solely to the vasodilatins almost invariably present in them.

Raw pancreas in diabetes mellitus. Neve (E. F.), Brit. M. J. (Lond.), 1926, **i**, 476.

A case report is given of a male, 16 years old. No blood sugar determinations were made. The urine showed 12% sugar at the time observation was begun. Allen treatment reduced the sugar to 4%. The continued use of insulin failed to maintain a low level. Administration of 2 to 3 oz. daily of raw pancreas gave sugar-free urine in 4 days. The patient was discharged a month later greatly improved.—C. I. Reed.

Influence of thymus extirpation on growth and the endocrine organs (Uber den Einfluss der Thymektomie auf den Gesamtorganismus

und auf die Drüsen mit innerer Sekretion insbesondere die Epiphyse und Hypophyse). Lindeberg (W.), Folia Neuropathologica Estoniana (Dorpat), 1924, **2**, 42-108.

The author carried out an extensive series of experiments (thymectomy) on dogs, cats, goats and pigs. Among other, the following results were noted: within the first month there appeared retardation in weight, dentrition was delayed, bone growth impeded, rhachitic changes were noted, there was muscle weakness and reflexes were augmented. Among the internal organs the most marked changes were noted in the brain, pineal, adrenals and liver. The pineal remained almost microscopic in size, the adrenals and liver were enlarged. The author regards these changes as proof of the endocrine function of the thymus. The literature is somewhat extensively reviewed.—R. G. H.

Blood sugar in status thymicolymphaticus. Maclean (Aubrey B.) & Sullivan (Ruth C.), Am. J. M. Sc. (Phila.), 1926, **171**, 659-669.

The blood sugar values in 3 cases of status thymicolymphaticus were found to be low (42, 52, 57 mg. per 100 cc.). In one case of suprarenal hemorrhage a blood sugar of 25 mg. per 100 cc. was found. In 6 cases of convulsions produced by conditions other than status thymicolymphaticus the blood sugars were normal or increases above normal. In 6 cases of enlarged thymus shown by roentgenograms the blood sugar in 5 cases was within normal limits and one very slightly decreased. In 6 cases where the blood sugar was determined within half an hour before death on patients with diseases other than status thymicolymphaticus, the values were normal or above normal. Acute suprarenal insufficiency is suggested as the immediate cause of sudden death in status thymicolymphaticus.—R. G. H.

Hyperthyroidism treated by x-rays. Barclay (A. E.) & Fellows (F. M.), Lancet (Lond.), 1926, **i**, 593.

A report is made of the results in 300 consecutive cases in private practice, 63.3% of which were pronounced cured and 25% definitely improved. Secondary conditions, such as cardiac disturbances, were of course not affected by the treatment. The smallest number of treatments effecting a cure was 6, the average 20. One-third of an erythema dose was given twice a week. Results are believed to be permanent. Of the total number of cases, 260 were females, 40 males. The greatest age incidence was between 30 and 50 years. Goiter was present in 75% of the cases, exophthalmos in 68%. The authors believe the basal metabolic rate determination inferior to observation and physical examination as a means of diagnosis. In 10 cases, diabetes was coincident.

—C. I. Reed.

Tumors of lateral aberrant thyroids. Billings (A. E.) & Paul (J. R.), Bull. of the Ayer Clinical Laboratory of the Pennsylvania Hospital, 1925, **9**, 27; Abst. Ann. Clin. Med., **4**, 688.

The authors report a case of papillary adenocarcinoma of a lateral aberrant thyroid and assemble 34 other cases from the literature. They find that these cases present a number of common features. In the great majority the tumor was found in the central portion of the neck beneath the sternomastoid muscle; in 6 cases it was localized close to the angle of the jaw, and in 6 in the supraclavicular fossa. In most of the cases the aberrant thyroid tissue nodules were multiple; in several instances they were noted on both sides of the neck. Twenty-six cases were in females, 6 in males, and no record of the sex was given in 3 instances. The average age at which surgical attention was drawn to the case was 34 for women and 42 for men. The youngest was a girl of 12 and the oldest a woman of 60. In 8 cases a definite malignant neoplasm was recorded as occurring in patients under the age of 30. The pathologic diagnoses varied from thyroid tissue, cystic thyroid to cystadenoma and adenocarcinomas. There is a pronounced tendency for lateral aberrant thyroid tissue to give rise to papillary adenoma and adenocarcinoma, as contrasted to the median or lingual aberrant thyroid which rarely becomes malignant.

A clinical and pathological study of fifty-five malignant neoplasms of the thyroid gland. Simpson (W. M.), Ann. Clin. Med. (Balt.), 1926, **4**, 643-667.

The results of a clinical and pathological study of 55 cases of thyroid malignancy are reported. Fifty were carcinomas, 5 were sarcomas. An incidence of 4.03% of thyroid malignancy among 1290 surgically removed, non-exophthalmic goiters examined histologically in the period from 1895 to 1925 was found. No instance of carcinomatous transformation of a purely hyperplastic (exophthalmic) goiter was encountered. In every case there was an associated preëxisting simple or adenomatous (endemic) goiter. Seventy-two per cent of the malignant thyroid neoplasms occurred in women. In three-fourths of the cases studied the malignant new growth appeared after the thirty-fifth year. Over 60% of this series were unsuspected carcinomas, the correct diagnosis being made only after histological examination. This fact emphasizes the necessity for pathological examination of all extirpated thyroid tissue. Clinical signs of hyperthyroidism were present in approximately one-half of the carcinoma cases. Every hard nodule in the thyroid gland of a person above thirty should be viewed with suspicion as regards malignancy. The frequency with which signs of hyperthyroidism are likewise present provides a second excellent reason for their surgical removal. The most important clinical evidence of early malignancy is a history of relatively rapid

increase in size and hardness in a previously quiescent or slowly growing goiter, particularly in a person over thirty years of age. Radiating neuralgic pain is a common early symptom. Metastasis is common in the advanced stages. The lungs are most often involved, while bone metastases are next in order of frequency. Many thyroid carcinomas are of unusually slow growth. The traditional five-year "danger period" is insufficient. Recurrence after five years is common. An unnecessarily ponderous classification of malignant neoplasms of the thyroid gland has arisen during the past half-century. Many of the terms used describe only minor histological variations, or are in themselves contradictory. It was found quite sufficient to divide carcinomas into three types, on the same basis used in classifying epithelial malignancy arising elsewhere in the body. This classification, based on the morphology and growth characteristics, is found to be in close agreement with the clinical aspects of thyroid malignancy. Medullary carcinomas make up 36% of the cases of malignant epithelial new growth studied. These neoplasms grow with the greatest rapidity, and usually terminate fatally in from one to four years. Recurrence and metastases are common. Those medullary carcinomas in which growth is restricted to a single adenoma offer a more encouraging outlook. Sixty per cent (30) of the carcinoma cases are designated adeno-carcinomas. Twenty-five (83%) of these definitely arose in adenomas, and it is quite probable that the remaining 5 had similar origin. Those adenocarcinomas which are restricted to a single adenoma (malignant adenomas) offer a more hopeful prognosis than those in which the malignancy is not thus limited. Not one of the "malignant adenomas" was diagnosed clinically. Most of these were removed for the relief of toxic symptoms. Scirrhous carcinoma was encountered but twice (4%). Neither case was diagnosed prior to operation. This type grows more slowly than the other type. The so-called "carcinosarcomas" are probably scirrhous carcinomas with an exuberant stroma reaction. Any form of thyroid malignancy may originate in aberrant thyroid tissue. Likewise, any pathological change possible in the thyroid gland, proper, may occur in the thyroid tissue of complex teratoid tumors of the ovary ("dermoid cysts"), sacrum, or elsewhere. We have encountered 2 carcinomas arising in the thyroid tissue of complex ovarian teratomas. Sarcomas of the thyroid gland conform in their growth characteristics to those arising elsewhere in the body. Sarcomas are but one-tenth as frequent as carcinomas in the material reported.

—R. G. H.